HEREDITY OF TWO VARIABLE CHARACTERS IN HABROBRACON

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INTRODUCTION. Review of work on defective venation in Lancaster (L) material.

In the parasitic wasp, *Habrobracon juglandis* (Ashmead), the fourth branch of the radius vein, r_4, has been found to show defects since part or all of it may be lacking in either or both wings. Temperature and other environmental conditions affect the character to a considerable extent but it has been found that when insects are reared at a uniform temperature of 30°C it is possible to estimate hereditary factors with a fair degree of accuracy. Results of studies made on about 95,000 wasps descended from a single female taken in Lancaster, Pennsylvania, (the L material), were discussed at length in a previous publication (WHITING, P. W. 1924). Inheritance is of the sex-linkoid type with exceptions due to patrocliny. (WHITING, P. W. 1921a, WHITING, ANNA R. 1925).

Various ratios of defectives occurred in the different cultures of the L material (WHITING, P. W. 1924, p. 12 ff.) and at least one "mutation" took place affecting this character. Very high percentages did not at first

appear but subsequently there were observed fraternities showing upwards of 90 percent defective. From these were isolated "high defective" stocks. (WHITING, P. W. 1924, p. 28 ff.). High defective acted as a unit, *d*, in heredity, the character showing in about 90–95 percent of males and 85–90 percent of females of the stocks and in about 45 percent of the males produced by heterozygous females. Variations in dominance of *D* over *d* occurred in the heterozygous females, *Dd*, so that 5 percent or more of them were defective. "Low defective" and "type" stocks were isolated but "medium defective" was lost. There was evidence that medium reappeared by "mutation" but this "mutant" strain was not retained. (WHITING, P. W. 1924, p. 33). Orange eye color acting as a unit occurred as a definite mutation (WHITING, P. W. 1921a) in the L material. Minor factors for defect causing occasional defectives in low defective stocks and acting as plus modifiers of *d* both for ratio and grade were postulated.

Stock 1, black, and stocks 5 and 8, orange, had normal venation, only 15 defectives occurring in 18,590 individuals, 0.0807 percent defectives.

Stocks 2, 6 and 7, orange, were called low defective stocks, showing about 0.325 percent defectives.

Stocks 3 and 10, orange, and stocks 4 and 9, black, were high defective. Cultures carrying *d* through twenty-eight generations involved fourteen crossings up to type stock alternated with parthenogenetic segregations (WHITING, P. W. 1924, p. 29 ff.). These failed to reduce percentage of defect significantly or permanently. Stocks 3 and 4 showed 94.9 percent defective in males and 90.2 percent defective in females and stocks 9 and 10, isolated at the end of this experiment, showed 94.4 percent defective in males and 89.9 percent defective in females. It may therefore be stated that high defective stocks differ from type stocks by only one significant factor for defective.

The method of origin of defective venation is of interest and is to be considered in the present paper. Its sudden appearance does not necessarily involve mutation in the true sense. Recombination, concentrating subliminal factors, may cause appearance of the character in a higher percentage than previously. It is supposed that there are numerous loci containing factors of different potencies acting in either a plus or a minus direction as regards the production of the character.

For convenience in reference the locus for orange eye may be regarded as in the first linkage group, Chromosome I, while the "main factor" for defective venation, since it segregates quite independently of orange (WHITING, P. W. 1924, p. 31), may be tentatively assumed to be in the second linkage group, Chromosome II. This factor or group of factors

acting as a unit may then be called d_{II95} since it produces about 95 percent defective males in pure stock under standard conditions. Its "normal" alternative, as in stocks 1, 5 and 8 may be called D_{II}. d_{II95} may have been formed by crossing over of d_{II} groups of lesser potency or by mutation of a single d_{II} unit.

Chromosome I also contains a lethal factor l_{Ia} causing premature initiation of metamorphosis of larvae into pupae, failure to spin cocoons and subsequent death of naked pupae. There were 59 crossovers to 243 straights or 19.5 percent crossovers. Lethal individuals could be classified as to eye color. (WHITING, P. W. 1921b).

Chromosome I also contains factors for defective venation acting as plus modifiers of d_{II95}. One of these of relatively low potency increased both percentage and mean grade of defectives in sons of $D_{II}d_{II95}$ females. It may be called d_{Ia}. It evidently occurs in stock 1 (WHITING, P. W. 1924, p. 47).

In addition there occurred by "mutation" in Chromosome I a plus modifier of greater potency which may be called d_{Ib}. This had the apparently anomalous effect of increasing the percentage of defectives from $D_{II}d_{II95}$ females while decreasing their mean grade. The explanation of this apparent contradiction is simply that $D_{II}d_{Ib}$ males are in part low grade defective while $D_{II}d_{Ia}$ males are practically all normal.

EXPERIMENTAL DATA

The Iowa City (I) material

Origin and differences from L material

In September 1922 a wild female was taken in Iowa City, Iowa. She produced 145 males and 186 females of normal wing venation. From these were bred the I material which showed several marked differences from the L material.

Fecundity was higher in that there were more offspring on the average per individual female. Either fertilizability of eggs or fertilizing capacity of sperm or both were higher as evidenced by higher female ratio from females mated within the strain and from the fact that reciprocal crosses with L showed higher female ratio than L alone. (WHITING, ANNA R. 1925).

I individuals are more excitable, fly more readily and are more easily lost by inexperienced students.

Crosses demonstrate differences in production of patrocliny. (WHITING, ANNA R. 1925).

Rare occurrence of thelytoky or the production of females from unfertilized eggs, never observed with certainty in the pure L material, has been demonstrated in I material and in crossbred material (LI).

Unpublished data show differences in number of antennal joints.

Different types of occasional freaks occur. There were recorded a very few, 15 males and 2 females, with thoraces showing a ventral split or unusually broadened. Five males had wings markedly reduced in size or venation and eleven had a prothoracic wing or wing sack on one side in addition to the normal primaries and secondaries. The last named type of aberration has never before occurred in studies of Habrobracon, nor have I any knowledge of its appearance in other insects. In eight of the eleven the structure was unexpanded, appearing at first as a sack filled with hemolymph. This later dried and shrivelled. In three it occurred on the right side, in five on the left. In the remaining three of the eleven the prothoracic wing was fully expanded, appearing on the right side in two, on the left side in one. When the wing was expanded the venation was similar to that of the normal primary. The point of origin of this sack or wing was just above but very close to the prothoracic leg.

"*Sooty*" *mesosternum*. The mesosterna, especially in the males, averaged much darker than the corresponding area in the L material. The character was called "sooty" and roughly graded as (1) yellow, with very little or no black pigment; (2) gray, with intermediate amount of black pigment; and (3) black, with mesosternum fully or almost fully black. L females are practically all yellow when bred at 30°C or above. I females are lighter than I males but darker than L males as is shown by the following record of material bred under standard conditions.

		YELLOW	GRAY	BLACK	TOTAL
I males	actual	744	486	7175	8405
	percent	8.85	5.78	85.36	
I females	actual	918	402	317	1637
	percent	56.08	24.56	19.36	
L males stocks 4, 5, 10	actual	471	36	20	527
	percent	89.37	6.83	3.79	

Defective venation. As regards defective venation, I material appeared slightly more defective than low defective L.

Approximately 12,678 males were graded, showing 71 or 0.560 percent defective, while 2315 females showed 17 or 0.738 percent defective. L low defective stocks averaged 0.325 percent defective. Breeding tests described below showed I to be very different from L as regards genetic aspects of defective.

Pure I material is being retained, designated as stock 11.

Results of crossing I and L material

Low defective crosses

L orange-eyed normal crossed with I. Color of sternum is not recorded in data presented in this section. Offspring of the original I female were crossed reciprocally with L stock 5 (normal wings, orange eyes).

Stock 5 male with I female gave all type, 63 males and 183 females, while the reciprocal cross produced 71 orange males and 85 type females. F_2 males are recorded in table 1; $I \times 5$, with reference to venation and eye color. The percentage of defectives, 1.80, is over three times as high as in pure I and many times as high as L stock 5.

In order to test more fully the production of defectives in the second generation from crosses of I and normal L, wasps from stock 8 and stock 11 were turned over in the spring of 1925 to ACHSA BEAN, a graduate studant at the UNIVERSITY OF MAINE. Stock 8 is an orange-eyed L stock of normal venation showing in the males but 3 defectives among 4336 studied (WHITING, P. W. 1924, p. 38). No change has been noted in this stock. Stock 11 as stated above is of pure I origin.

F_2 males counted by MISS BEAN are recorded in table 1; 8×11. The ratio of defectives, 8.14 percent, is greatly in excess of that for the earlier experiment, 1.80 percent. Both exceed that of the pure I males, 0.560 percent, and of normal L males, 0.0807 percent. These differences may be attributed to chance selection of material used in the crosses.

L orange-eyed low defective crossed with I. An orange male from stock 6 (low defective L, 0.325 percent defective) was crossed with a black I female. Classification was made on the basis of eye color, venation and mesosternum, the latter being called simply yellow or sooty. F_1 showed only normal venation and black eyes, 26 sooty males and 77 yellow females F_2 males are recorded in table 1; 1×6.

Black sooty of mixed origin (LI) crossed with L orange-eyed normal. A sooty male from the F_2 generation just considered was mated to stock 5

female resulting in 25 orange males and 39 black females. All but one of each sex had yellow sterna. F_2 males are recorded in table 1; 5♀ × F_2♂ (I×6).

Three defective sooty males from combinations of I material with stocks 5, 6 and 10 were crossed with stock 5 females. There resulted males; sooty 2, orange 104, orange sooty 10, females; type 98, sooty 9. (It may be noted that the two patroclinous males are sooty as well as black-eyed showing paternal character while the majority of the regular males as well as the females have yellow sterna.) F_2 males are recorded in table 1; 5♀ × ♂♂ from I, 5, 6, 10.

In the I×5 series there occurred a male with eyes that were mosaics of black and orange (Freak 154). He bred like a black when crossed to stock 5 females and the F_2 generation from his daughters showed that he transmitted sooty. F_2 males are recorded in table 1; 5 ♀ × mosaic eye ♂ (I×5).

Summary and discussion; failure of linkage of orange, sooty, and defective. The various crosses discussed above involved pure L, stock 5, 6 or 8, on the one side. On the other were black-eyed sooty wasps, either of pure I origin or derived from I crossed with various orange L stocks. Hence the character black-eye and the "main factor" for sooty-sternum came into the cross from one side, orange-eye and yellow-sternum from the other. Among the 4306 F_2 males classified with reference to orange and sooty, there are 49.4 percent recombinations, and hence no evidence of linkage.

As regards defective venation it is impossible to tell the origin of the factors involved. No "main factor" seems to be present since both parents occasionally show defective and the factors may recombine. On the average the I material is somewhat more defective than the L; hence coupling with black or with sooty is to be expected if any linkage exists.

Among the total blacks are 4.032 percent defective and among the total orange 3.105 percent defective. The difference which is slightly less than three times its probable error may possibly indicate the presence of a minor factor for defect, d_1, comparable with such as have been previously shown (WHITING, P. W. 1924, p. 45 ff).

Among the total sooty are 2.506 percent defective and among the total yellow are 3.033 percent defective. This indicates that there is no apparent linkage between the low defective factors and sooty.

Frequencies and ratios are given in table 1.

TABLE 1

F_2 males from black-eyed sooty crossed with orange-eyed yellow, showing segregating relationships of orange, sooty and low defective.

PARENTAL CROSS	STERNUM	BLACK Normal	BLACK Def.	ORANGE Normal	ORANGE Def.	TOTAL	PERCENT DEF.	PERCENT ORANGE	PERCENT SOOTY
I × 5		336	8	374	5	723	1.80	51.04	
8 × 11		512	38	473	49	1072	8.14	48.69	
I × 6	yellow	107	4	91	2	458	4.15	46.29	55.46
	sooty	125	10	116	3				
5♀ × F_2♂ (I × 6)	yellow	36	1	24	1	142	4.23	47.89	56.34
	sooty	35	2	41	2				
5♀ × ♂♂ from I, 5, 6, 10	yellow	334	23	351	13	1439	3.96	50.73	49.90
	sooty	334	18	363	3				
5♀ × mosaic eye ♂ (I × 5)	yellow	445	7	466	7	2267	1.15	49.18	59.20
	sooty	687	13	633	9				
Totals	yellow	922	35	932	23	4306	2.74	49.35	55.60
	sooty	1181	43	1153	17				
	Totals	2951	124	2932	94	6101	3.59	49.60	

Effects of crossing, selection and inbreeding on defective and sooty. The I by 5 series. Origin of stocks 12, 13, 14.

Some of the defective males of the F_2 generation from the original I by 5 cross were mated back to I females and from the progeny lines were continued, the I by 5 series. Selection was made roughly for defective and sooty, both orange and black eye color being retained. Three stocks were isolated all with very dark sterna.

Stock 12 rarely shows defective. Percentages of defectives in the males of the various generations in its ancestry tracing backwards were 2.0, 11.7, 4.6, 1.2, 3.5, 0, 2.4, 0. No definite counts to determine ratio of defectives in pure bred stock 12 have been made, but ANNA R. WHITING in studies of patrocliny recorded venation character in orange males from stock 12 females crossed to black males. They totaled 1968 including 38 or 1.930 percent defective. Stock 12 is therefore a low defective stock.

Relations of stock 12 in production of patrocliny have been discussed by ANNA R. WHITING (1925).

Stock 13 with orange eyes and stock 14 with black eyes were isolated from highly defective lines. Stock 13 had percentages of defective males in various generations as follows: 68.6, 80.0, 51.5, 50.0, 19.5, 33.4, 58.6, 36.9, 15.4, 11.7, 4.6. It joined the common line with stock 12 in the P_4 generation of the latter in which there were 1.2 percent defective males. Stock 14 had 50.0, 41.6, 31.3, and joined the common line with stock 13 in the P_5 generation of the latter in which there were 19.5 percent defective males. U. A. HAUBER (1925) has selected stock 14 for still higher grade of defect and made combinations with d_{II95}, the "main factor" for defective which occurred by mutation in the L material. Thus stocks have been obtained showing practically 100 percent defective.

Stocks 12, 13 and 14 have been continued as "pure" stocks.

Both "sooty" and "defective" showed considerable variation in the I by 5 series. "Sooty" ranged from jet black to clear yellow which is typical of stock 5 as of all L material bred at 30°C or over. The average depth of pigmentation in some lines as in stocks 12, 13 and 14, exceeded considerably pure I material. For the entire series the males showed sterna yellow 162, gray 194, and black 5739, out of a total of 6095. The females showed sterna yellow 1115, gray 1417, black 1123 out of a total of 3655. Percentages of various grades of sooty for I and for the entire I by 5 series are brought together for comparison as follows:

I males.....	yellow 8.85	gray 5.78	black 85.36	Total 8405
I×5 "	" 2.66	" 3.18	" 94 16	" 6095
I females....	" 56.08	" 24.56	" 19.36	" 1637
I×5 "	" 30 51	" 38.76	" 30.73	" 3655

The I by 5 series is therefore darker with respect to mesosternum color, not only in certain of its branches but on the average, and in both males and females.

Linkage of high defective and sooty

Black-eyed males with sooty sternum and r_4 defective due to low defective factors were crossed to stock 10 females. There resulted 13 black patroclinous males, 170 orange males and 145 black females. F_2 males were graded as yellow or sooty with respect to sternum color, and also classified with respect to orange and defective. Results are recorded in table 2, group A.

A sooty male with normal venation and black eyes occurring in F_2 from stock 6 male crossed with I female was crossed to stock 10 female. There resulted 53 orange-eyed males, 10 normal and 43 defective, graded as yellow 29, gray 20, and black 4. These were typical stock 10 males.

There were also produced 131 black-eyed females of which 3 were defective. All were graded with respect to sterna showing 110 yellow, 17 gray and 4 black. F_2 males are recorded in table 2, group B.

An orange-eyed sooty male from I material bred up to stocks 5 and 6 was crossed to stock 4 female. There were produced males; type 1, defective 66, and females; type 126, defective 2, no sooty appearing in F_1. F_2 males from two virgin F_1 females are recorded in table 2, group C.

An orange defective (grade 8) male derived from combinations of I with stocks 6 and 10 was crossed with stock 11 female. There were produced black males, 1 yellow and 49 sooty, black females, 87 yellow and 8 sooty. F_2 males are recorded in table 2, group D.

TABLE 2

F_1 males from crosses involving orange, sooty and high defective, showing segregating relationships.

GROUP	PARENTAL CROSS	EYE COLOR	TYPE	SOOTY	DEFECTIVE	SOOTY DEFECTIVE	TOTAL		ORANGE	DEFECTIVE	SOOTY	CROSSOVERS (?)
A	10♀ ($oS_{11}d_{11\,s}$)×	Black	11	72	62	53						
	♂$O_{911s}D_{11}d_x$	Orange	9	117	61	44		actual	231	220	286	117
		Total	20	189	123	97	429	percent	53.84	51.05	66.67	27.27
B	10♀ ($oS_{11}d_{11s}$)×	Black	57	370	286	26						
	♂$O_{911s}D_{11}$	Orange	58	394	296	30		actual	778	638	820	171
		Total	115	764	582	56	1517	percent	51.29	42.06	54.05	11.27
C	4♀ ($OS_{11}d_{11\,s}$)×	Black	17	51	45	2						
	♂$os_{11}D_{11}$	Orange	18	48	55	1		actual	122	103	102	38
		Total	35	99	100	3	237	percent	51.48	43.46	43.04	16.03
D	11♀ ($O_{911s}D_{11}d_x$)×	Black	7	57	48	11						
	♂$oS_{11}d_{11\,s}$	Orange	4	57	51	14		actual	126	124	139	36
		Total	11	114	99	25	249	percent	50.60	49.80	55.82	14.46
	Total	Black	92	550	441	92						
		Orange	89	616	463	89		actual	1257	1085	1347	362
		Total	181	1166	904	181	2432	percent	51.60	44.61	55.39	14.88

The four summaries of F_2 males from defective by sooty are totaled with respect to these two characters at the bottom of table 2. It may be seen that there is clear evidence of linkage for the recombinations, crossovers (?), are but 14.88 percent of the total. This may be regarded as a

"repulsion" series but since both characters are variable there is no evidence that any of the 362 apparent crossovers are genetic. Moreover any of the 2070 apparent straights may be genetic crossovers. Crossing over could be tested by breeding the apparent crossovers to a parent stock. This test would be complicated by recombination of supernumerary factors both for sooty and for defective.

The "main factor" or group of factors for sooty, showing linkage with d_{II95}, may be called s_{II}. Supernumerary factors for sooty of unknown linkage groups may be called s_X, and similarly supernumerary factors for defective may be called d_X.

Referring to the totals in table 2 we find that out of 2432 F_2 males, orange is very close to the expected 50 percent. There is a marked deficiency of defectives, doubtless due to the character overlapping with normal, despite the presence of supernumerary factors, d_X, in some of the groups. There is a marked excess of sooty, probably due to supernumerary factors, s_X.

Formulae for the parental crosses of the various groups are shown in the second column. Females are from stock 10, ($oS_{II}d_{II95}$) orange yellow defective, stock 4 ($OS_{II}d_{II95}$) black yellow defective, and stock 11 (Os_{II} S_X D_{II} d_X) black sooty sooty-supernumerary normal defective-supernumerary. Males are of diverse composition but always different from females with reference to o, s_{II} and d_{II95}. As regards supernumerary factors, s_X and d_X, their formulae are hypothetical in part although based on origin, character, and progeny.

Orange offspring do not depart significantly from expectation in any of the groups.

Defective is in excess of expectation in groups A and D, s_X coming from the male in A, from the female in D. (It must be noted that expectation is well below 50 percent due to overlapping, and that a ratio of 50 percent or over indicates supernumerary factors, d_X, if temperature is standard, 30°C).

Sooty is in excess in groups A, B, and D, coming from the males in A and B, from the female in D. In groups C and D the males have been freed of supernumerary factors possessed by I material, stock 11, by crossing with L material.

Variation in ratios of crossovers (?) might be correlated with presence of supernumerary factors but this cannot be done satisfactorily in the present instance, because of differences in standard of grading. It would also have been more satisfactory if parental males had been of pure stock.

GENERAL SUMMARY AND CONCLUSIONS

Hereditary factors governing defective venation (breaks in the fourth branch of the radius, r_4) and sooty mesosternum are discussed. Both characters show continuous variation and are much affected by environmental factors, temperature in particular.

In certain stocks of Lancaster material (L) there occurs a factor, d_{II95}, producing about 95 percent defectives among the males. Other L stocks are known as type, (less than one-tenth percent defective), and low defective (less than one percent defective). Total Iowa City material (I) recorded, 12, 678 males and 2315 females, showed less than one percent defective.

F_2 generations from crosses of I material with type or low defective L or with individuals of mixed origin (LI) show no evidence of linkage between sooty and defective.

Certain of these F_2 generations indicate recombinations of factors increasing ratio of defectives. This theory is strengthened by the great increase of defectives in certain branches of a series derived from I crossed with normal L material, I by 5 series. Stocks 13 and 14, derived from the I by 5 series show very high ratios of defectives, over 50 percent, without the presence of d_{II95}.

Sooty mesosternum occurs in I material of which 8405 males and 1637 females were bred and graded in this respect. Males of either L or I material tend to be darker than their corresponding females but I females are darker than L males.

Recombination of factors for sooty is shown by the mesosterna of the I by 5 series and by stocks 12, 13 and 14 derived from it. Stocks 12, 13 and 14 have very black mesosterna in both males and females. Of the I by 5 series 6095 males and 3655 females were graded, averaging darker than the I males and females respectively.

F_2 males from high defective, d_{II95}, crossed with sooty were type 181, defective 904, sooty 1166 and defective sooty 181. Since characters are variable this is not to be considered a correct gametic ratio. It does, however, demonstrate linkage of the main factor for defective, d_{II95}, with the main factor for sooty which may therefore be called S_{II}. This linkage taken in connection with failure of linkage between low defective and sooty possessing s_{II} indicates that the low defective under consideration was not in Chromosome II.

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CHROMOSOME BEHAVIOR IN A CROSS BETWEEN WHEAT AND RYE[1]

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INTRODUCTION

A number of investigators have succeeded in crossing wheat and rye, but only with great difficulty and after many trials. There is a variety of wheat, however, which crosses readily with rye, as high a proportion of pollinations proving successful as in crosses between any two varieties of wheat. This variety I have secured through the kindness of MR. W. O. BACKHOUSE (1916) of Buenos Aires. By using it as the female parent a large F_1 and a very small F_2 have been produced. From these and from back crosses a great many plants of later generations have been raised. This material, particularly the F_1, has been studied cytologically and the results are reported in the following pages.

KIHARA (1919) and SAX (1922) have shown that the cereals furnish an excellent example of polyploidy. The basic chromosome number is 7. Among whe ats diploid, tetraploid, and hexaploid species are known, the familiar bread wheats having 21 haploid chromosomes. In rye the haploid number is always 7 so far as known. The hybrids between 14— and 21—chromosome species of wheat show at the heterotypic division 14 bivalent and 7 univalent chromosomes. Those between 7—and 14—chromosome species show 7 bivalents and 7 univalents. It is concluded that in these

[1] This work was carried on with the aid of a grant from the Research Council of Canada.

cases each of the chromosomes of the species with the smaller number finds a mate among the chromosomes of the other species, leaving 7 of the latter unmated. No hybrid between a 7—and a 21-—chromosome wheat has been studied cytologically.

KIHARA (1924) has given some results of an examination of a single F_1, hybrid between a *vulgare* wheat and winter rye. At the heterotypic division 0, 1, 2 or 3 bivalents were formed, and the number of univalents varied correspondingly between 28 and 22. Some of the univalents divided equationally while others failed to do so. At the homeotypic division a variable number of lagging undivided chromosomes appeared. No material of later generations was available.

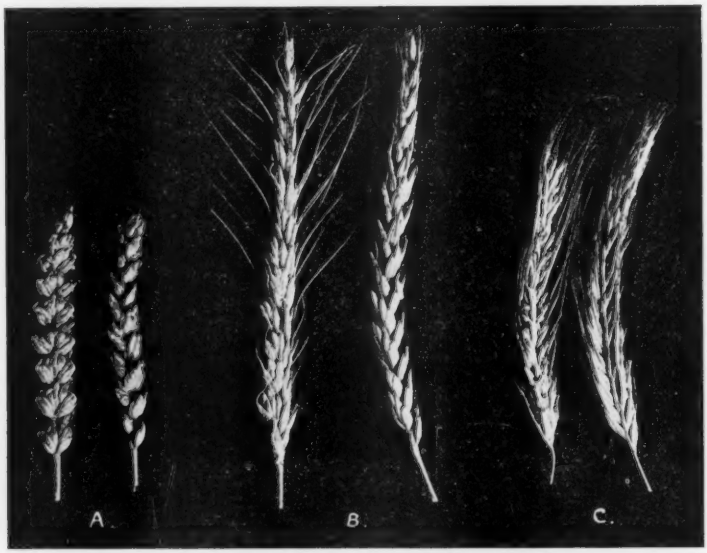


FIGURE 1.—A, wheat parent; B, F_1; C, rye parent.

CHARACTERS OF PARENTS AND HYBRIDS

The wheat used (figure 1, A.) is typical *Triticum vulgare* var, *albidum* Korn. It is a spring wheat of Chinese origin. It shows no peculiarities which would lead one to expect unusual behavior in crossing with either rye or other wheats. The only respects in which it resembles rye a little more closely than do average *vulgare* wheats is in the somewhat more pronounced keel of the outer glume, and the longer, sharper beak of this glume. But many other *vulgare* varieties have these structures as well

developed. A marked characteristic of the spike is the sharply incurved, short awns. It behaves like an ordinary variety of *T. vulgare* in crosses with other *vulgare* varieties and with other species of Triticum, except that in certain instances the species crosses are more successful when it serves as one parent.

The rye parent (figure 1,C) chiefly used is a spring type known locally as Prolific. It resembles the variety Petkus. Other sorts were found to cross as readily.

The hybrid (figure 1,B) is very vigorous, at least equalling and often exceeding even the rye parent. Eighteen characters which distinguish the parents were examined. The F_1 is like rye in 2, like wheat in 6, and intermediate in the remaining 10. In regard to 2 of these 10 it is almost exactly intermediate, but resembles rye more than wheat in 3 and wheat more than rye in 5. There is thus a strong preponderance of wheat characters, a fact which may be due to the larger number of wheat chromosomes.

The few F_2 were very variable but for the most part the wheat characters were still more pronounced. A few more rye-like conditions were present and rarely rye characters appeared which were absent in F_1. It is proposed to deal later with the botanical characters in detail and their relation to chromosome conditions.

METHODS

Most of the material was studied by means of smears in iron-aceto-carmine. Many preparations were made of fresh material but the majority were of material fixed in Carnoy's solution and preserved in 70 percent alcohol. The latter method is superior in certain respects; the differentiation is better, and the making of slides and their examination need not be hurried during the short flowering season but may be done at any later time. It is inferior in that the pollen mother-cells do not remain in cylinders but separate, and, when they become flattened after the first division lie in such a position that it is difficult to get polar views. For the material of this study the best reagent was produced by adding 2 drops of a saturated solution of ferric hydrate in 45 percent acetic acid, to 50 cc of a half-saturated solution of carmine in 45 percent acetic acid. The preparations may easily be sealed with asphaltum and then keep excellently for a month or two, gradually becoming darker.

A considerable amount of material was also sectioned in paraffin and stained in Haidenhain's haematoxylin.

CHROMOSOMES OF THE PARENTS

The chromosomes of the wheat are illustrated in figure 2,A and B.[1] They are similar in number (21) and form to those of *vulgare* varieties in general. They are short, thick, and shaped like bent dumb-bells, or rings,

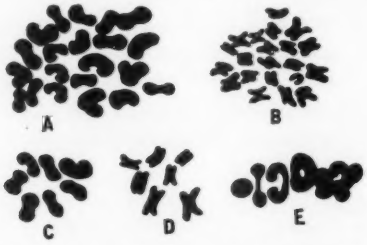


FIGURE 2.—Heterotypic chromosomes of parents. A, wheat metaphase; B, wheat anaphase; C, rye metaphase polar view; D, rye anaphase; E, rye metaphase side view.

or U's or V's with enlarged ends. The equatorial plate is regular and rarely is there even a slight lagging. In anaphase each one splits longitudinally and the ends diverge,so that they appear as 2 sickles or U's attached by the middle.

The variety of rye used has 7 haploid chromosomes (figure 2, C, D, E,) shaped like those of wheat and behaving similarly. Rye furnishes particularly favorable material since numerous stages are found in one spike, the stamens are very long and easily removed, and the chromosomes few and large.

FIRST REDUCTION DIVISION OF F_1

At the heterotypic division 28 chromosomes make their appearance in the great majority of cases (figure 3A). This is the sum of the haploid numbers of the two parents (rye 7, wheat 21). Evidently no mating of chromosomes occurs in these cases and all that appear are univalent. Occasionally counts of 27 and 26 are made and rarely 25. In these cases shorter, stouter bivalents can usually be distinguished and can be seen dividing in their characteristic fashion (figure 5). Evidently mating of 1 or 2 pairs may take place occasionally and of 3 pairs rarely. In a series of counts made to determine the proportions in which the different numbers

[1] Figure 2 is drawn from paraffin sections. A camera lucida, Zeiss 2 mm apochromatic objective and 25×ocular were used. The magnification is reduced 2/3 in reproduction to 1500. Figure 6 was made similarly but with 15x ocular (×950). All other drawings were made from smears in iron-aceto-carmine with 3 mm dry apochromatic objective and 25× ocular (×900).

occur it was found that 49 cells had no bivalents, 8 had 1 and 2 had 2 bivalents.

The individual chromosomes are quite different in form from the normal heterotypic ones of the parents. They are longer, thinner and more curved. Most of them are sickle-shaped, some U- or V-shaped. On the other hand they are not like the vegetative chromosomes nor those of the second reduction division. The latter are still longer and slenderer and almost invariably much curved into V's and I's (figure 8). Why the unmated univalent chromosomes of the heterotypic division should differ from vegetative or homeotypic ones is not clear. The occasional bivalents closely resemble those of the parents, being short, stout rings or dumb-bells.

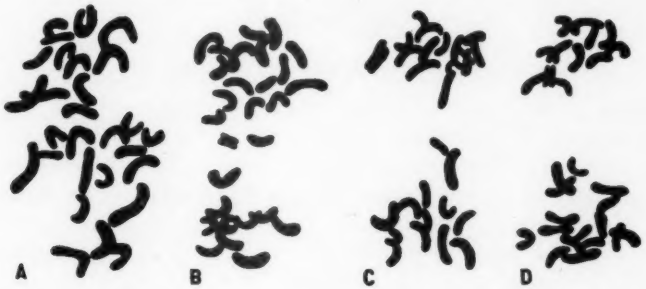


FIGURE 3.—Heterotypic division of F_1, 28 univalent chromosomes separating into 2 groups without division of individuals.

A striking feature of the heterotypic division is the entire absence of an equatorial plate. The shape of the entire group of chromosomes changes from a sphere to an ellipsoid and this becomes longer and narrower by the movement of the chromosomes towards the ends of the cell. The whole group gradually separates into two sub-groups without division of the individual chromosomes (figure 3). There is in fact nothing resembling an equatorial plate stage. The chromosomes simply scatter towards the ends in a long, frequently narrow band. The appearance is therefore very irregular and untidy. Commonly those which reach the end first are reforming a nucleus long before the last ones have reached the end. The separation is usually into approximately equal groups. Usually each group contains 12 to 16, though counts as low as 9 have been made. Apparently the number which goes to either end is determined by the chance disposition of the individuals on either side of the middle line where the equatorial plate would normally form.

Usually before the ends are reached some of the chromosomes show a longitudinal split like that which is seen in the anaphase of the heterotypic division of the parents. This results in the appearance of two parallel rods or 2 slightly curved rods attached at the middle and diverging towards the ends. This appearance is quite different from that of a dividing bivalent. The halves of the latter long remain attached at one end and become much

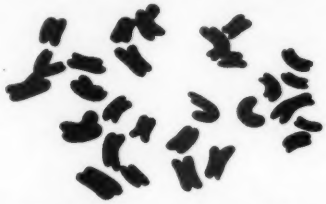


FIGURE 4.—Heterotypic division of F_1 28 splitting univalents.

drawn-out. In a considerable proportion of cases nearly all the chromosomes show the univalent split even before the two groups have separated (figure 4). One may find in many cases almost the whole 28 split and still showing no sign of segregation into 2 groups. The two halves of a split chromosome do not normally go to opposite ends but move along together to one end, like the similar chromosomes during anaphase of a

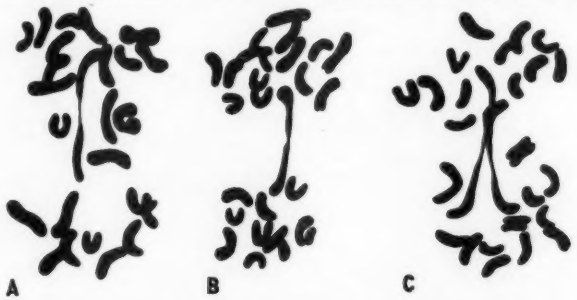


FIGURE 5.—Heterotypic division of F_1 showing dividing bivalents.

normal heterotypic division. Occasionally the halves of one which happens to lie on the equator may move to opposite ends. The usual failure of the halves of a univalent to move to opposite ends is in contrast to the behavior of the univalents in species crosses within the genus *Triticum*. In those crosses the univalents divide at the heterotypic division and wander undivided to one end or the other at the homeotypic. In the Aegilops-wheat hybrids on the other hand SAX (1924) finds that the univalents

usually fail to divide at the first division, though they may occasionally do so.

It has been mentioned that in a percentage of cases only 27 or 26 chromosomes appear and that 1 or 2 bivalents may then be recognizable. These divide in characteristic fashion, the two halves remaining attached at one end for a long time and becoming much elongated (figure 5). The connecting portion becomes drawn out into a thin strand. The bivalents may usually be seen dividing after the univalents have separated at least partially into 2 groups, but they may divide earlier.

Commonly some of the chromosomes fuse and form a daughter nucleus before others have reached the end (figure 6). Late-comers often fuse with each other and form additional nuclei, as many as 4 being seen in some cases. Their size is very variable. Even when several nuclei are present isolated lagging chromosomes are usually to be seen. Commonly 2 or more nuclei formed in this way eventually fuse, and consequently at this stage

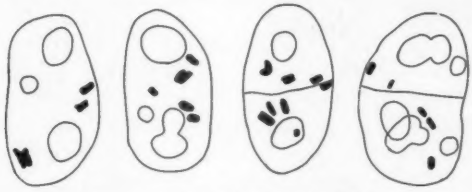


FIGURE 6.—End of heterotypic division.

the nuclei are frequently irregular in outline. Usually one or more of the additional nuclei become cut off by walls, so that previous to the homeotypic division there are frequently 3 or 4 or more cells, some much smaller than others.

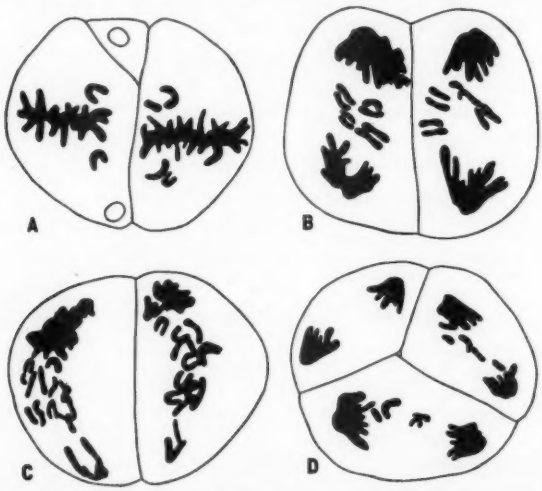
The prophases of this division may now be considered in the light of the later events. In spite of the facts that no mating of chromosomes usually occurs, and that never more than a few chromosomes are involved in mating, the prophasic events appear to be nearly normal. Though only a preliminary examination has been made of the details of prophases, enough has been done to ascertain that only one contraction occurs. Parallelism of chromatin strands is also evident. If further work shows that this is usual it will have a decisive effect on the parasynaptic interpretation of the similar pairing in the pure species, since in this hybrid little or no mating occurs.

The events at the heterotypic division in Kihara's single hybrid differed in several respects from those just described. Bivalents were present in a larger proportion of the cells. An irregular equatorial plate appeared on

which many univalents frequently failed to become arranged. Though actual numbers are not mentioned, apparently a much larger proportion of the univalents divided equationally than in my material.

SECOND REDUCTION DIVISION OF F_1

The homeotypic division is usually much more regular than the heterotypic. A more or less definite equatorial plate is usually formed (figure 7, A). Sometimes all the chromosomes are found in the plate; sometimes 1 or 2 are seen off it; usually a fairly large number fail to reach the plate before the majority have divided. Sometimes no equatorial plate is formed at all (figure 7, C.), and then a very irregular division figure is seen. At



FIGURES 7.—Homeotypic division. A, typical metaphase side view; B, lagging chromosomes splitting; C, very irregular division; D, 3 nearly equal cells dividing.

this division the chromosomes are long, slender, and much curved or bent. All or nearly all divide (figure 7, B.). Occasionally in individual chromosomes there is no indication of division. These are presumably the occasional univalents which divide at the first division.

The number of chromosomes varies within wide limits. This variation is a result of the irregular scattering at the heterotypic division, very different numbers often reaching the two ends. It also depends on the fact that frequently more than one nucleus is formed and becomes enclosed in a cell. In those cases in which irregularities are least evident the number

varies from 12 to 18. Such cases are represented in figure 8. In the supernumerary cells numbers as low as 2 may be counted.

The extra nuclei usually divide at the same time as the others. One may then frequently find 3 to 5 cells of varying size within the old wall of the mother cell all dividing at the same time, some regularly and some very irregularly. Often small nuclei which are in cells along with large ones fail to divide.

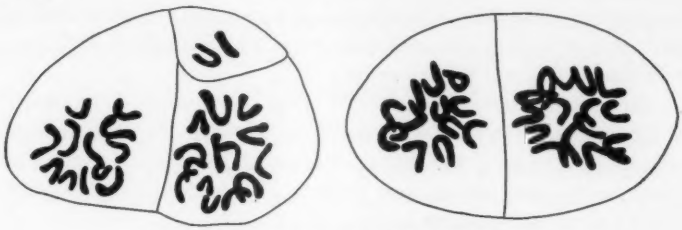


FIGURE 8.—Homeotypic division, polar views.

As in the heterotypic division the chromosomes which first reach the end may form a new nucleus before the later ones arrive. In this way 2 or more nuclei are aften formed and individual chromosomes may remain outside all of them. Often these isolated chromosomes enlarge and form very small nuclei; often they appear to degenerate. There is frequently a fusion of the 2 or more nuclei in a cell.

TETRADS AND POLLEN

Owing to the irregularities at both divisions the resulting "tetrads" usually present a very abnormal appearance (figure 9). While 4 cells may

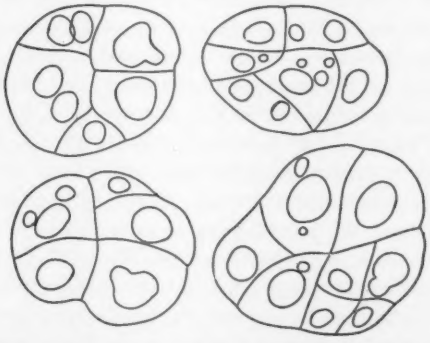


FIGURE 9.—Typical "tetrads" showing extra cells and nuclei and fusion of latter.

commonly be seen in the group, any number from 2 to 10 may occur, "tetrads" of 5, 6 and 7 being very common. The numbers seen in a series of counts were as follows:

Number of cells	2	3	4	5	6	7	8	9
Number of cases	2	4	29	36	26	14	3	2

It is possible that those with 2 were delayed and would still divide but they were seen at a stage when many of the others were already showing degeneration. The two cells were much larger than those of other "tetrads." A number of those with 4 included 2 very small ones. The variation of size of the cells is very great.

Usually some of the cells show more than one nucleus. The number of additional nuclei decreases with maturity, showing that fusion occurs, a point also indicated by the frequently irregular shapes. In many cases, however, more than one nucleus remains in a cell until it shows signs of the death of the protoplasm.

The young pollen grains show indications of abortion while still in the tetrads, and before there is any evidence of their separation. The protoplasm shrinks and takes on a fibrous appearance. The small cells are the first to show these changes.

The mature pollen grains are nearly all empty and shrunken. In a count of 500 in fuchsin and lactic acid only 4 showed well stained protoplasm and this was thin and vacuolate.

CHROMOSOME CONDITIONS IN LATER GENERATIONS

Owing to the scarcity of F_2 plants and the desire to obtain as many seeds as possible no F_2 material was examined. An F_3 plant showed 17 bivalents and 2 univalents, and an F_4 plant of another line showed 17 chromosomes among which no univalents could be distinguished nor any lagging chromosomes. These plants had several distinctively rye characters though for the most part they were like wheat.

From several seeds obtained on unprotected F_1 a large number of descendants have been raised. These were probably the result of natural back crossing with nearby wheat plants, since they were wheat-like in most of their characters, and since the stigmas of F_1 remain fresh and expanded for long periods. Several lines do show some rye characters, however, and it is possible that they were not the result of back crosses but that the more numerous wheat chromosomes dominate the few rye ones. Several plants from these lines were examined. None had fewer than 21 chromosomes at the heterotypic division, some had exactly 21, and some had more. The highest number observed was 25 and this included several

univalents. In all these cases good equatorial plates were formed. In one case 3 plants from one family all showed 22 chromosomes and no univalents could be detected either by form or lagging. It appears that a line has been started with a higher chromosome number than either original parent had.

A plant produced by artificially back-crossing an F_1 with the wheat parent also had more than 21 chromosomes. It showed at the heterotypic division 21 bivalents and 3 univalents. The behavior was similar to that in species hybrids in wheat.

DISCUSSION

It is evident from the behavior at the first reduction division that each chromosome of *T. vulgare* has a special partner with which it mates in the pure species. Since *T. vulgare* is hexaploid the chromosome set of the ancestor of modern wheats has presumably been triplicated in some way in prehistoric times. Consequently there should be 7 groups of 6 similar chromosomes in the somatic cells and 7 groups of 3 similar ones in the reproductive cells. It might be expected, then, that in pure *T. vulgare* any one of the 6 might mate with any other one. This random mating occurs, according to SAX (1922), in recent examples of chromosome-multiplication in Oenothera, Datura and Primula. In regard to Oenothera this statement is denied by GATES (1924).

In wheat the genetic evidence with multiple factors indicates that the assortment is not random. The behavior at the heterotypic division in these wheat-rye hybrids confirms that conclusion. None of the wheat chromosomes usually finds a mate though there should be 3 of each kind, even if the rye chromosomes are disregarded. The usual absence of mating shows that in pure wheat each chromosome has its own particular partner. As SAX (1922) has suggested the chromosomes may have differentiated so much since the original triplication that random mating is no longer possible. Since all wheat species were distinct in prehistoric times (PERCIVAL 1921) there has been sufficient time for the differentiation.

The occasional presence of 1 or 2 bivalents may, however, indicate that some of them are able to mate occasionally with chromosomes which are not their normal partners, if those partners are absent. Of course a bivalent may also be the result of mating between a rye and a wheat chromosome. But it would seem more probable that chromosomes which have been duplicated within the genus *Triticum* would mate with each other than with chromosomes from another genus. Furthermore, WINGE (1924) has given evidence that in pure *vulgare* wheat a chromosome may occasion-

ally mate with one which is not its normal partner, and that one of the results is a speltoid mutation. If such a mating may occur when the partner is present it is still more likely to occur when the partner is absent. But the usual absence of bivalents in the wheat-rye hybrids, and their very small number when present, show that wheat chromosomes usually remain faithful even to their absent mates.

COLLINS and MANN (1923) have shown that in species hybrids in Crepis the behavior is similar. In the F_1 of *Crepis capillaris* (haploid 3) and *C. setosa* (haploid 4) no mating occurs and the 7 univalent chromosomes pass undivided to the poles. In the hybrid between *C. setosa* and *C. biennis* (haploid 20) the *biennis* chromosomes all mate in 10 pairs leaving the *setosa* ones unmated. Though the wheat-rye cross is much wider botanically the chromosome behavior is intermediate between those two Crepis types since an occasional mating occurs.

The only other genus cross which has been carefully studied cytologically is the one between *T. vulgare* and *Aegilops cylindrica*. SAX (1924) finds the behavior in this much more regular. *Aegilops* has 14 haploid chromosomes. In the F_1 about 7 bivalents and 21 univalents are usually formed. Supernumerary nuclei and cells are very rare. SAX suggests that 7 Aegilops chromosomes mate with 7 wheat ones and that 14 wheat and 7 Aegilops ones remain unpaired. In some cases he was not able to find the full 7 bivalents nor in many cases the full 21 univalents.

In view of their behavior in Aegilops-wheat hybrids, in rye-wheat hybrids, and in species hybrids within the genus Triticum, it would appear that the chromosome relationships in wheat and its relatives would be as follows:

	Sets of seven chromosomes				
	A	*B*	*C*	*D*	*E*
T. vulgare	x	x	x		
T. durum, dicoccum, etc.	x	x			
T. monococum	x				
Aegilops			x	x	
Secale					x

The three sets of 7 haploid chromosomes of *T. vulgare* are designated A, B and C. Two of these mate with the 2 sets of the emmer types of wheat which may then be designated A and B. One of these latter mates with the single set of einkorn which may therefore be designated A. Set C will then be distinctively *vulgare*. The set in Aegilops which mates with

one set in *T. vulgare* will probably be C since, as Percival has pointed out, the important characters which distinguish the *vulgare* from the emmer type are all found in Aegilops. If this is true the other set in Aegilops must be incompatible with either A or B; otherwise it would mate with one of them and 14 bivalents and 7 univalents would be found. This different set may then be designated D. (The existence of a distinct set would have to be assumed even if the Aegilops set which mates with *vulgare* chromosomes be considered A or B). Therefore two 14-chromosome types, Aegilops and emmer wheat, have all their chromosomes incompatible. Nevertheless TSCHERMAK (1913) was able to cross Aegilops with all species of wheat. Since there is no botanical or genetic reason for thinking that the rye set is similar to Aegilops set, and since it does not mate with A or B or C it must be considered different from all the others. There is not sufficient evidence to decide where all these differentiations occurred or how the multiples were produced in different lines. But further combined genetic and cytological work on species and genus crosses should throw light on the problem.

It is possible that a definite set of 7 Aegilops chromosomes does not always mate with a set of 7 *vulgare* ones. Occasionally SAX could find no more than 5 bivalents and frequently less than 21 univalents, so that there may be a variable amount of mating. This may correspond to the variable though smaller amount of mating in wheat-rye hybrids. The latter is probably between wheat chromosomes only.

This work shows, however, that great caution must be exercised in drawing conclusions regarding relationships from ease of crossing or chromosome numbers. This particular variety of wheat crosses with rye as easily as with any other species of wheat, much more easily than with some species, and quite as easily as any variety of *vulgare* crosses with any other variety. It is not difficult to get 90 percent of pollinated flowers setting seed, while JESENKO (1913), working with a number of *vulgare* wheats, secured only 35 seeds from 6129 flowers pollinated with rye (and 12 of those from 1 head). Einkorn wheat which has the same number of chromosomes as rye (7) cannot be crossed with rye, while this particular *vulgare* variety with 3 times the number may be crossed readily. Similarly it has been found almost impossible to cross a certain variety of *T. dicoccum* (14 chromosomes) known as Khapli, with *T. vulgare* though numerous attempts have been made, owing to its high rust-resistance. Very few seeds are obtained, and these few either fail to germinate or produce F_1 which fail to reach maturity. Other varieties of *T. dicoccum* cross readily with *T. vulgare*, and the F_1 shows a high degree of fertility. It is evident

that a large number of varieties must be used in species crosses before final conclusions can be drawn.

It is also evident that conclusions regarding economic results must not be too hastily drawn from a consideration of chromosome numbers. It appears that some of the chief advances in wheat breeding depend on the crossing of different species—a procedure which presents great difficulties because of the differences in chromosome numbers. But it is clear that very different results may be expected when different varieties of the same species are crossed with a different species. In the present example we have a large number of lines in late generations with a preponderance of wheat characters but also certain definite rye characters. Judging from chromosome numbers and experience with other attempts to cross wheat and rye, this would be impossible.

The rare production of seed on selfed F_1 plants and the chromosome numbers of the F_3 and F_4 examined show that viable plants are occasionally produced without the full complement of wheat chromosomes. The botanical characters of these plants also show that they carry rye chromosomes. The absence of some *vulgare* chromosomes has not tended in these cases to produce *emmer* characters.

The considerable proportion plants of later generation resulting from back-crosses, which had more than 21 chromosomes is of interest. In all these cases there were at least 21 bivalents; in most of them there were 21 bivalents and a number of univalents; in one case a stable condition with 22 bivalents appears to have been reached. It would be of great interest both theoretical and economic to produce a stable race of wheat with more chromosomes than any now in existence. It is not easy to see how plants with more than 21 chromosomes have arisen immediately following a back cross, unless the events in the female may occasionally be different from those in the male. If the chromosomes in the megaspore mother cell did not scatter undivided as in the microspore mother cells, or if rarely the division is so irregular that only 4 or 5 chromosomes go to one end, and all the others to the opposite end, the result might be achieved, though even in the last instance 21 must be wheat chromosomes since 21 bivalents are formed in the offspring. If, as in the lily and numerous other angiosperms (COULTER and CHAMBERLAIN 1903) the megaspore mother cell could develop directly into an embryo-sac without forming megaspores, then the fusion of 2 nuclei from opposite ends of the female gametophyte for endosperm formation would restore approximately the full haploid complement of rye and wheat chromosomes. Such a fusion nucleus might be expected after fertilization to grow into an embryo

instead of endosperm, since its chromosome complement is like that of an egg. According to WATKINS (1925), however, 4 megaspores are produced in wheat and only one functions. What happens in rye is unknown.

It is not so difficult to account for the appearance of plants with more than 21 chromosomes in later generations. If an F_1 female gamete with 18 chromosomes (a number seen at the homeotypic division in pollen mother cells) including 3 rye ones, is fertilized by a male gamete with 21, the heterotypic division in the offspring would be expected to show 15 bivalent wheat chromosomes, 6 univalent wheat and 3 rye. Occasional gametes may then be expected to have 24 chromosomes (21 wheat and 3 rye) which being fertilized by 21 chromosome gametes from the same plant or pure wheat would give the desired condition.

SUMMARY

A certain variety of *vulgare* wheat crosses as easily with rye as with any other *vulgare* variety and more easily than with other species of wheat. The proportion of successful pollinations is as high as in crossing any two varieties of wheat. The F_1 is very vigorous and not quite completely sterile.

The wheat used has the typical 21 haploid chromosomes of *Triticum vulgare* and the rye has the typical 7 of Secale.

At the first reduction division in F_1, 28 univalent chromosomes usually appear. These move without dividing to opposite poles, nothing like an equatorial plate being formed and the division figure appearing highly irregular. Often 1 or 2 and rarely 3 bivalents appear and divide in characteristic fashion. Supernumerary nuclei are formed from lagging chromosomes; these may fuse with the principal nucleus, or may become enclosed in cells of their own, or may remain unfused in the same cell as the principal nucleus.

The prophases of this division appear from preliminary observations to show only one contraction. Some parallelism of chromatin strands is evident though not expected on the parasynaptic interpretation.

The second division is usually much more regular than the first, but may be quite as irregular. An equatorial plate is usually formed on which nearly all the chromosomes become oriented and divide, though lagging chromosomes are common. "Tetrads" usually have from 4 to 8 cells but may have more or fewer.

From back-crosses plants with more than 21 chromosomes have been secured.

In tetraploid and hexaploid species of wheat each chromosome has a single mate; random mating amongst duplicated chromosomes does not occur normally, but may occur abnormally. It is probable that there is among wheat chromosomes no mate for any rye chromosome.

Caution is necessary in drawing conclusions concerning relationships from ease of crossing or chromosome conditions.

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MUTATIONAL MOSAIC COAT PATTERNS OF THE GUINEA PIG

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INTRODUCTION

Most of the guinea pigs which one sees have a mosaic coat pattern of some sort. The piebalds have a pattern of colored spots on a white ground, the tortoiseshells have a pattern of dark spots on a yellow ground and the tricolors have both yellow and dark colored spots of some sort on a white ground. It has been suggested (EYSTER 1924) that these patterns may be due to somatic mutations of an unstable gene, such as has been demonstrated in variegated corn. There certainly appears to be something akin to mutation as far as cell lineages are concerned. On the other hand, the more or less orderly relationship to the parts of the body indicates that the pattern is determined primarily by developmental processes of the type responsible for ordinary regional differentiation. The genes for piebald (*s*) and tortoiseshell (e^p) are simple recessives, seemingly as stable as any other genes in the germ-plasm. THE BUREAU OF ANIMAL INDUSTRY has maintained a number of inbred strains of guinea pigs since 1906. One of these families (No. 34) was tortoiseshell (SSe^pe^p) throughout its history. All of the others have bred consistently tricolor (sse^pe^p) except that in one of them (No. 13) true breeding red-white piebalds (*ssee*) segregated out in

one line near the beginning. Some 35,000 guinea pigs have been recorded in these lines and from crosses between them, but there is not the slightest evidence that the piebald factor has ever mutated to its dominant allelomorph (S) or that the tortoiseshell factor has mutated either to dominant true-breeding black (E) or to the recessive true breeding red (e). Black-white and red-white bicolors appear not infrequently but breeding tests have indicated that these differ genetically in no respect from their tricolor relatives, their behavior contrasting sharply with that of blacks (EE) and reds (ee) obtained from other sources. With this type of mosaic pattern we shall have little further concern in this paper.

There are five sets of allelomorphs in guinea pigs which normally produce a uniform change in color throughout the coat except that the degree of effect may show an entirely regular regional differentiation. Mosaics in the effects of these factors are extremely uncommon, but when they do occur they presumably indicate some sort of a mutational or chromosomal change in the region affected. The purpose of the present paper is to give an account of the seven possible cases of this sort which the writers have noticed among over 40,000 guinea pigs which they have recorded.

The record of every guinea pig born in the experiments includes a sketch of the pattern made in a rubber stamp outline, similar to the patterns shown in the figures. These drawings have been made for the most part by the junior author, who has been on the lookout for any irregularities of the kinds to be described. The senior author has gone over all of the animals born each week for the purpose of recording the intensities of the various colors. Other opportunities for observation occur at the time of weaning and at times when weights are taken. Doubtless a few inconspicuous cases have been overlooked, especially in animals born dead. We feel safe in saying, however, that such mosaics are extremely uncommon in guinea pigs.

THE COLOR FACTORS OF GUINEA PIGS

The color factors which are known to have been present in this stock are given below. The black-eyed golden agouti variety with all 7 dominant factors is taken as the type, as having the factorial composition of the wild Cavia cutleri in these respects. In this variety each hair is black with a "red" (*ochraceous tawny*) subterminal band. This band is wider and lighter in color on the belly, which thus has a yellow appearance.

S, s	s = piebald (white areas). Incomplete dominance.
E, e^t, e.	e^t = tortoiseshell (red areas).
	e = self red

A, a^r, a	a^r	= narrow red ticking, ticked belly
	a	= self black
C,c^k,c^d,c^r,c^a,	c^k	= slight dilution of black, marked dilution of red
	c^d	= marked dilution of both black and red
	c^r	= slight dilution of black, red replaced by white, eyes red
	c^a	= black reduced to sooty white, red replaced by white, eyes pink (albino)

In all cases, intensification of black, but slight fading of yellow on exposure to cold and with age. Incomplete dominance in compounds of lower members of this series.

F,f.	f	= dilution of red only (except in presence of p), effect increasing markedly with age.
B,b.	b	= brown in place of black; dark red eyes. No effect on red.
P,p.	p	= pale sepia in place of black; pink eyes. No effect on red.

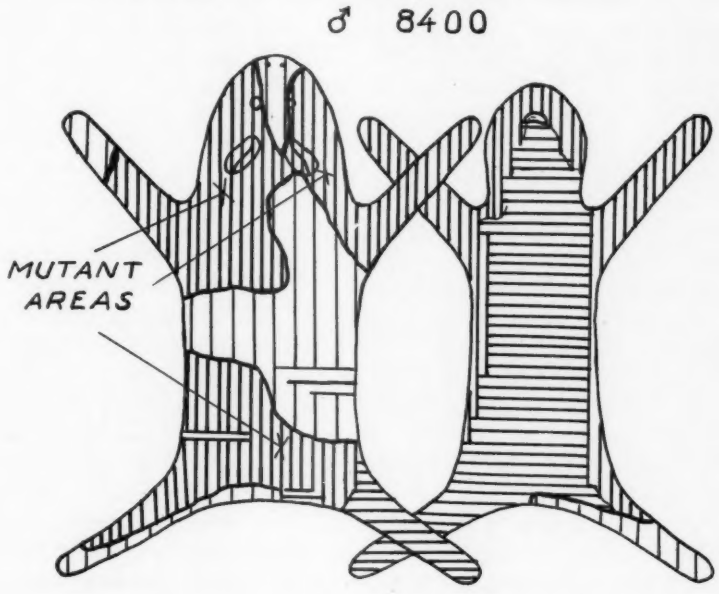


FIGURE 1.

The coat pattern of ♂8400. Vertical lines represent the agouti and horizontal lines the red or yellow of his tortoiseshell pattern. The heavily shaded portion represents the mutational intense portion (golden agouti and red) while the light shading represents the normal yellow agouti and yellow.

As already noted mutational mosaics could not easily be detected in piebalds (ss) or tortoiseshells ($e^p e^p$) because of their naturally mosaic patterns. The mosaics to be described apparently involved A in one case, the C series in one case certainly and possibly in two others, F in two cases

and *P* in one case. In only one case is there evidence that the germ-plasm was affected. We will describe this case first.

A SOMATIC AND GERMINAL MOSAIC ($Cc^d+c^dc^d$)

Male 8400 attracted attention at birth (December 23, 1918) through having a mosaic pattern of red agouti and yellow agouti. There were two separate red agouti regions as shown by the heavy bars in figure 1. One of these included the head about the eyes and ears, the shoulders and right fore leg. The other included both flanks and the right hind leg. Thus, the nose, left fore foot, a belt around the middle (extending forward on the right shoulder) and finally the left hind leg and left side of the rump were yellow agouti. The pattern was sufficiently striking at birth but became less easy to recognize later, largely, apparently, because of a relatively greater amount of fading of the red agouti parts. On brushing back the hair it may be seen that the black as well as the red is more intense in the red agouti areas. The differences are such as would be expected between an intense agouti (C-) and a dilute agouti (c^dc^d)[1]. Approximately 60 percent of the coat is red agouti dorsally. The entire belly appears yellow, but as there is little difference in intensity between "intense" and homozygous dilutes in this region, it is probable that the mutant patches extend over part of it.

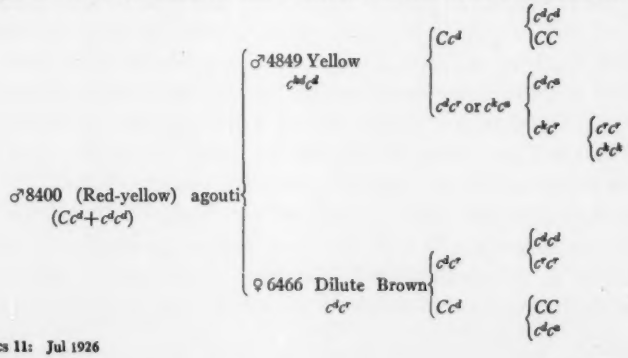
This animal attracted special attention from the first because of the fact that both parents were dilutes and thus should not transmit the dominant intense coloration which he clearly showed. The sire (♂4849) was a self yellow of grade 8, believed to be c^dc^d with c^kc^d as a possibility. He was mated twice. His first mating was with a black-cream tortoise-shell necessarily c^kc^r from her parentage. The progeny consisted of 3 with yellow in the fur (c^kc^d) and one with cream (c^dc^r). His second mating was with a dilute brown (♀6466) which later was proved by breeding tests to be c^dc^r. This mating produced 9 with yellow in the fur (c^dc^d) and 4 with cream (c^dc^r) in addition to the mosaic male (♂8400) showing both red and yellow which was thus assumed to be genetically c^dc^d. The color of the sire, together with his breeding record of 17 dilute young, from dilute females, omitting the intense dilute mosaic, agree in indicting that he was a homozygous dilute. His own sire, however, was an intense (agouti-red-white) necessarily Cc^d as coming from a cross between a yellow male (c^dc^d) presented by Professor W. E. CASTLE and a golden agouti tricolor female of an inbred family (No. 39) which had always been CC. His dam was a cream that might have been either c^dc^r or c^kc^a as coming from a mat-

[1] The colors of the different albino series compounds are described in detail elsewhere (Wright, 1925). The intensity of yellow is the best criterion. Using compound symbols, C– is typically red of grade 10 or 11, $c^{kd}c^{kd}$ yellow of grade 6 to 8, $c^{kd}c^{ra}$ cream of grade 4 or 5 and $c^{ra}c^{ra}$ pure white.

ing between a cream, c^dc^a, presented by Professor CASTLE and a black-cream c^kc^r. from a mating between a red-eyed brown (c^rc^r) presented by Professor CASTLE and a black-yellow from an inbred family (No. 18) homozygous c^kc^k in the branch concerned. The brothers and sisters of ♂4849 were 3 with red (*C*-), 1 with yellow ($c^{kd}c^d$) and 3 with cream (c^dc^{ra}) in harmony with the formula assigned his parents.

The possibility that ♂4849 may have transmitted *C* from his father must of course be borne in mind, but the assignment of formula Cc^d to him raises difficulties in accounting for his own color and his failure to sire any unequivocal intense among 18 young, without assisting much in accounting for his mosaic son.

Going back to the dam of the mosaic, ♀6466 was a brown, a color in which the effects of the *C* series are not easily distinguished. She was mated three times, however, with results that make it certain that she was c^dc^r. Her first mating was with ♂4849, in which, as above noted, she produced 9 young with yellow (c^dc^d), 4 with cream (c^dc^r) in addition to the mosaic. Her second mating was with an albino male (c^ac^a) made especially as a test. She produced two sepia-cream young clearly c^dc^a from their color, 2 sepias proved to be c^dc^a by breeding tests, and 2 red-eyed sepias, clearly c^rc^a from coat and eye color. Her third mating was with a red-eyed dark sepia (c^rc^r). She produced 9 black-eyed dilutes (c^dc^r) and 9 of the appearance of homozygous red-eyed dilutes (c^rc^r). She thus had 38 graded young to 23 of which including the mosaic she supplied c^d and to 15 of which she supplied c^r. It is perhaps worthy of record that she produced her 16th litter at the advanced age of five years and one month. She died a month later. Like ♂4849, she had an intense parent but in view of her extensive breeding record in which it was proved conclusively that she transmitted both c^d and c^r, the possibility that she transmitted *C* seems negligible. The formulae of the ancestors of the mosaic are shown below:



Before leaving the ancestry, it may be well to state that a careful investigation was made at once to determine the possibility that the parentage of the mosaic had been incorrectly assigned. He was born in his parents' second litter, with two litter mates, a sepia-yellow (c^dc^d) and a sepia-cream (c^dc^r). There was no evidence that any intense guinea pigs had been in the pen in which he was found except his reputed parents for seven months. Female 6466 undoubtedly had a litter at this time. Breeding tests showed that the mosaic transmitted *e* and *b*, factors necessarily transmitted by ♂4849 (*ee*) and ♀6466 (*bb*) but which could have come from only a small number of other animals in the stock, descended from a few animals presented by Professor CASTLE. It happened that all but one of these other matings produced a litter within two months before or two months after. The one exception had been sterile for a year and could have produced the mosaic only through a double mutation. There is thus exceptionally complete evidence in this case that there was no mistake in parentage.

The genetic constitution of the mosaic was tested by matings with albinos (c^ac^a) red-eyed dilutes (c^rc^a) and one dark-eyed sepia (c^dc^a). The matings with albinos and red eyes were desirable since the distinction between the red of Cc^{ra} and the cream of c^dc^{ra} is more striking than that between the red of Cc^d and the yellow of c^dc^d. In order to obtain as many young as possible he was moved in a ten-week cycle through three pens, in each of which there were usually two or three females.

The litters are shown in table 1 in chronological order. The first litter contained a golden agouti (intense) in addition to a cream agouti, thus proving that he transmitted intensity. The next litter confirmed this conclusion. Altogether, he produced 79 intense young and 149 dilutes. The expectation on the basis of his parentage and the appearance of the lighter areas in his coat was 100 percent dilutes. If, however, he were a heterozygous intense (Cc^d) the expectation would be 114 intense to 114 dilutes. The actual result is, of course, wholly out of harmony with the first hypothesis and deviates from the second by 35 ± 5.0, or seven times the probable error. The chance of obtaining such a deviation in this direction is about 1 in a million. Extensive experiments with the albino series have given no other significant departures from the expected ratios. Thus it seems safe to conclude that ♂ 8400 did not breed like a normal Cc^d. The obvious suggestion is that he was a mosaic germinally as well as somatically. If his germinal epithelium were 70 percent Cc^d and 30 percent c^dc^d, the observed ratio would be obtained. The dilute progeny and

TABLE 1

The breeding record of ♂8400. All of the females with which he was mated were as indicated albinos (c^ac^a) or red eyed dilutes (c^rc^a) with one exception, No. 8084, a dark eyed dilute (c^dc^a).

DATE OF BIRTH	DAM No.	DAM formula	INTENSE	DILUTE
1919 Sept. 2	215	c^ac^a	1	1
Nov. 5	215	c^ac^a	1	1
Nov. 30	8084	c^dc^a	2	2
Dec. 17	752	c^rc^a	1	1
Dec. 29	9154	c^rc^a		1
1920 Jan. 23	1317	c^rc^a		2
Jan. 28	8203	c^ac^a	1	3
Feb. 23	752	c^rc^a		1
Mar. 9	215	c^ac^a	1	1
Mar. 31	1317	c^rc^a	1	1
Apr. 9	8084	c^dc^a	1	3
Apr. 29	752	c^rc^a		3
May 29	215	c^ac^a	1	2
June 9	8203	c^ac^a	1	3
July 3	9154	c^rc^a		1
July 6	752	c^rc^a		3
Aug. 4	1317	c^rc^a	1	2
Aug. 7	215	c^ac^a		1
Sept. 4	4496	c^rc^a		1
Sept. 9	4497	c^rc^a		1
Sept. 10	752	c^rc^a		2
Sept. 10	9154	c^rc^a		2
Oct. 18	2	c^rc^a	2	2
Oct. 30	8203	c^ac^a	1	3
Nov. 14	4497	c^rc^a	3	1
Nov. 18	752	c^rc^a		1
Dec. 6	215	c^ac^a	1	
Dec. 15	1317	c^rc^a	1	1
Dec. 24	2	c^rc^a	1	4
Dec. 26	4496	c^rc^a	2	1
1921 Jan. 23	752	c^rc^a	1	1
Mar. 27	7523	c^ac^a	1	1
Mar. 28	4497	c^rc^a	1	2
Apr. 1	752	c^rc^a	1	2
Apr. 20	1317	c^rc^a	2	1
Apr. 23	215	c^ac^a	1	1
May 7	7970	c^ac^a		2
May 8	7928	c^ac^a		3
May 13	8203	c^ac^a		1
June 7	752	c^rc^a		2
July 5	1317	c^rc^a		1
July 13	7928	c^ac^a	5	1
Aug. 5	7533	c^ac^a	1	3
Aug. 13	752	c^rc^a	2	2
Sept. 17	8203	c^ac^a	3	1
1921 Sept. 20	7928	c^ac^a	1	2
Oct. 14	7533	c^ac^a	3	1
Oct. 19	752	c^rc^a	1	2
Nov. 1	1317	c^rc^a	1	2
Nov. 25	8203	c^ac^a	1	4
1922 Feb. 8	7928	c^ac^a	1	2
Feb. 26	752	c^rc^a		2
Mar. 5	7533	c^ac^a		3
Mar. 31	1305	c^ac^a	2	2
May 20	752	c^rc^a		2
Sept. 5	7928	c^ac^a	1	1
Sept. 26	8203	c^ac^a		1
Sept. 27	752	c^rc^a	2	2
Oct. 10	14963	c^ac^a		1
Nov. 15	7045	c^ac^a	1	1
Nov. 22	14789	c^ac^a		2
Dec. 14	7928	c^ac^a		3
1923 Jan. 7	752	c^rc^a	1	2
Jan. 8	14963	c^ac^a		1
Feb. 14	14962	c^ac^a	1	1
Feb. 20	7928	c^ac^a	1	1
Mar. 14	752	c^rc^a	1	3
Mar. 14	14963	c^ac^a		
Mar. 20	16355	c^ac^a		2
Apr. 7	7045	c^ac^a	2	1
May 19	14963	c^ac^a	1	2
May 26	16355	c^ac^a	1	1
July 4	14962	c^ac^a		2
July 9	7928	c^ac^a		2
Aug. 1	16355	c^ac^a	2	1
Aug. 13	17608	c^ac^a	1	2
Aug. 25	7045	c^ac^a	1	4
Sept. 11	7928	c^ac^a	3	2
Sept. 20	14963	c^ac^a		3
Oct. 1	752	c^rc^a	1*	
Oct. 5	16355	c^ac^a	1	2
Nov. 26	14962	c^ac^a		3
Dec. 9	752	c^rc^a	5	1
Dec. 9	14963	c^ac^a		
Dec. 12	16355	c^ac^a	1	3
1924 Jan. 18	17608	c^ac^a	1	2
Total			79	149

*A "black-white" born dead is not included, as it might have been either Cc^a or c^dc^r.

their descendants showed grades of sepia which indicated that the dilution factor which he transmitted was always c^d rather than c^b.

There is some indication that he did not produce the same ratio of intense to dilute at all times. Up to the end of 1919, his record was 5 intense to 6 dilute, giving no indication that he was breeding differently from Cc^d. During the next nine months, however, he produced only 7 intense to 32 dilute. The total results, summarized to this point, deviated from equality by 5.4 times the probable error and were presented by the senior author at the meeting of the American Society of Naturalists in 1920 as fairly conclusive evidence of an aberrant ratio. There followed, however, a period of thirteen months in which his record 34 intense to 42 dilute showed a much less impressive departure from equality. Indeed, in one four-month period, July to October, 1921, an excess of intense (16:13) was produced. During the next seventeen months there was again a marked deficiency of intense: 13 intense to 38 dilute. The final ten months with 20 intense to 31 dilute, seemed to show another shift toward equality. These variations in ratio can only be taken as suggestive, however, since on comparing with the expected figures on the basis of a constant proportion (34.7 percent) of intense, we obtain a value of P of .125 for the χ^2 test. Thus irregularities as great as those observed would be expected to occur one time in eight purely by chance. Another test can be obtained by comparing the record of 34 intense to 42 dilute produced in 13 months with the entire remaining record. The result is a contrast of 44.7 ± 3.8, with 29.6 ± 2.5 percent, a difference of 15.1 ± 4.6, about 3.3 times the probable error which might be expected to occur about 1 time in 35 by chance. Neither of these tests is quite fair in that they involve a selection of the periods for comparison. Nevertheless, there is a distinct suggestion that the proportion of germ cells carrying intense and dilute varied from time to time. This could be explained on the hypothesis that the germinal epithelium was mosaic and that the relative activity of different portions varied. A belated attempt of an experimental test of this point was made by removing the left testis (October 18, 1923). Only one litter was sired following the operation. This litter included 1 intense to 2 dilute, proving that both C and c^d were transmitted by the germinal epithelium of the right testis. Male 8400 died, (December 30 1925) at the rather advanced age of seven years, but produced no young since the litter referred to above, although mated with eight females for much of the time.

A number of his progeny have been tested by mating with albinos or red eyes, or with each other. There is nothing unusual in their records. Six intense young mated with albinos or red eyes have produced 46 intense

to 59 albinos or red eyes, a ratio which does not depart significantly from equality. Two matings between intense young have produced a total of 24 intense to 8 albinos, exact expectation. One homozygous dilute son, from the one mating in which ♂8400 was paired with a dilute, has produced 9 typical dilute young in a mating with an albino. Two heterozygous dilute sons mated with albinos have produced 8 dilute to 5 albinos where equality is the expectation. A mating between two dilutes produced two dilutes. The intense grandchildren of ♂8400 appear to be exactly like ordinary intense animals.

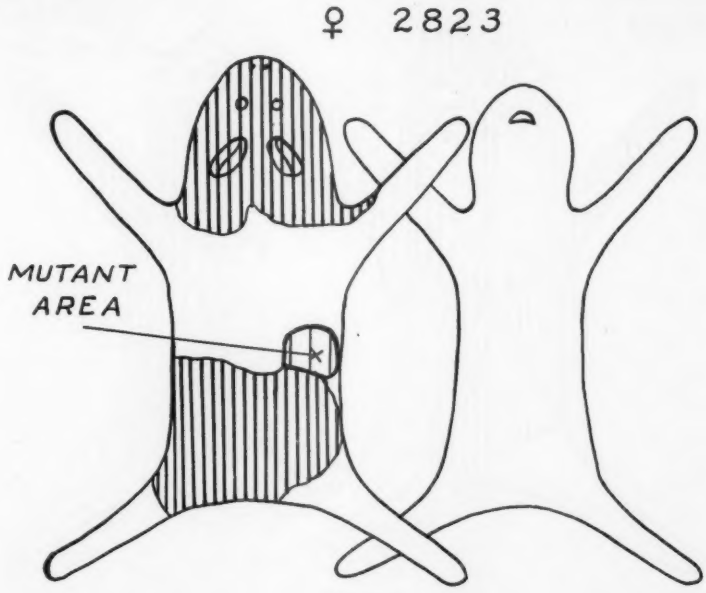


FIGURE 2.

The dorsal coat pattern of ♀ 2823. The heavy vertical lines represent black and the light vertical lines sepia. There was a red spot and another sepia spot on the belly, not shown in the early record. The mutational significance of the sepia spots is very doubtful.

The simplest explanation of this case is that ♂8400 was initially $c^d c^d$ but that a mutation to the dominant allelomorph *C* occurred in a cell which was ancestral both to part of the soma and part of the germinal epithelium.

It must be confessed, however, that it is difficult to entirely dismiss the thought that one of the intense grandparents somehow transmitted his

intensity through the apparently dilute sire or dam. The writers, however, have been unable to devise any explanation along this line which does not raise more difficulties than it solves.

TWO INTENSE-DILUTE MOSAICS

Two intense dilute mosaics may be noted very briefly. One of these (♀2823) was born in one of the inbred families (No. 38) homozygous in

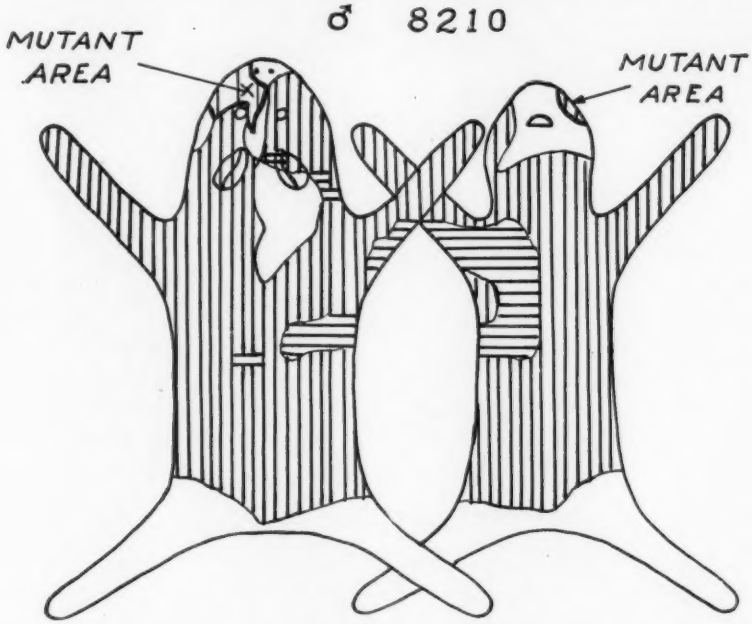


FIGURE 3.
The coat pattern of ♂ 8210 showing black (heavy vertical lines), sepia (light vertical lines) and red (horizontal lines). The mutational significance of the sepia spot is very doubtful.

all color factors. This guinea pig had four sharply distinct colors in the coat, black, sepia, red and white (figure 2). Incidentally, she had the peculiarity of lacking both eyeballs, a trait which appeared sporadically in family 38. Her 11 progeny were all blacks without any of the abnormal sepia color. A grandson, however, ♂8210, born December 3, 1918, was somewhat like her in his quadri-colored coat. He had a sepia patch on his nose contrasting sharply with the surrounding black as well as with his

red spots (figure 3). He came from a mating which brought together four different inbred families (13, 39, 34 and 38) from all of which he could have received only the intensity factor *C* in the albino series, except for a possibility of receiving albinism, c^a, from family 13. There was no evidence from the numerous progeny of his grandparents and parents or from his own progeny, which included 3 normal blacks from a mating with his dam, and 4 normal blacks from a mating with his sister, that he received or transmitted anything but *C* in the albino series, or that other factors pro-

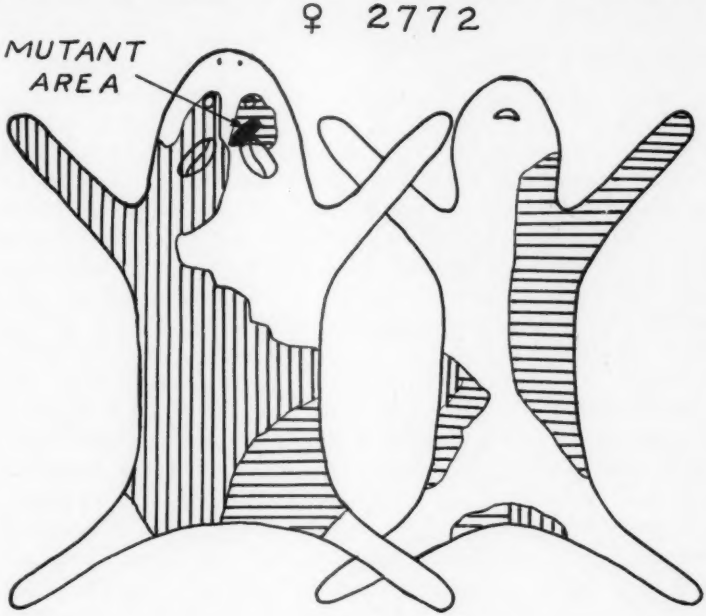


FIGURE 4.
The coat pattern of ♀ 2772 showing golden agouti (vertical lines), black (solid) and red (horizontal lines).

ducing dilution of black were present. A possible explanation is that the sepia spots represented modified brindled spots. Tortoiseshells usually show not only clear-cut black and red areas but brindled areas in which black and red hairs are intermingled. In these areas the black is usually much reduced in intensity. A spot composed exclusively of such hairs, that is, dilute black unassociated with red hairs, would have much the appearance of the observed spots on ♀2823 and ♂8210. If this is the

explanation, the mosaic pattern has no mutational significance, being simply a spot due to factor e^p, in which both animals were homozygous. It could be assumed that a peculiar combination of modifying factors gave these spots their unusual appearance.

AN AGOUTI-BLACK MOSAIC ($AA+aa$)

We will next consider the case of an agouti-black-red-white quadricolor, ♀2772, born November 24, 1919, in stock *B*, the control stock for

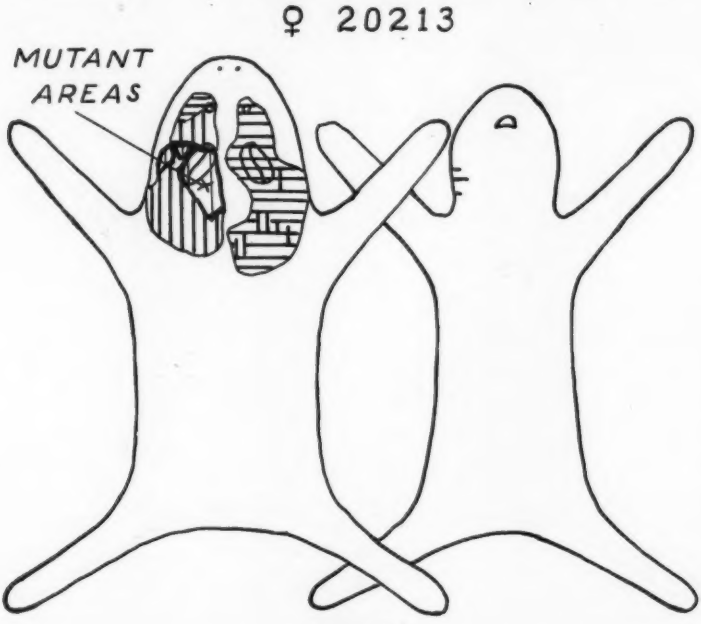


FIGURE 5.
The coat pattern of ♀ 20213 showing golden agouti (heavy vertical lines), cream agouti (light vertical lines) and red (horizontal lines).

the inbred families. This female had a clear-cut black spot in the fur in front of her right ear, surrounded by red (figure 4). She is the only agouti guinea pig on which the writers have ever noticed as much as a solid black hair. Her parents, grandparents and great-grandparents were all agoutis, except for one red-white bicolored great-grandparent, probably an agouti genetically. Three out of the 16 great-great-grandparents, however, were black. ♀2772 was one of fourteen agoutis produced by her parents. She

herself produced only 14 agoutis in a mating with a black male. Two of her daughters were mated with black males and produced 23 agoutis to 25 blacks, in addition to red-whites and albinos in which the agouti factor can produce no visible effect.

The ancestry and breeding record of ♀2772 make it reasonably certain that she was a homozygous agouti (*AA*) genetically. If the black spot was due to the ordinary recessive black factor (*a*) both genes must have mutated or been lost. The possibility remains, of course, that a mutation to an unknown dominant black occurred in some other locus.

TWO INTENSE-DILUTE MOSAICS (*Ff*+*ff*)

The next case is that of a golden agouti tricolor, ♀20213, born in the control stock *B*, September 4, 1923. In this case a much broken yellow agouti spot was noted on the left side of the head (figure 5). This spot changed later to a very pale cream agouti, markedly in contrast with the surrounding golden agouti. The black base of these cream agouti hairs was of full intensity however. These effects all indicated the kind of dilution due to factor *f* rather than the dilution of the albino series.

The sire was an albino, whose sire came from mating *B*211 which (with its parent matings) is the only known source of factor *f*. The paternal great-granddam of ♀20313, curiously enough, was ♀2772, the agouti-black mosaic discussed above. The dam of ♀20213 was a red-white from golden-agouti tricolor parents and grandparents. The parents produced 23 golden-agouti tricolors, 7 black-red-white, 2 red-white in addition to the mosaic. This result indicates that the dam was *CCFF*, the sire being probably $c^a c^a Ff$.

Female 20213 was herself tested by mating with a black-yellow-white male, known to be $Cc^a ff$ from his parentage. She produced 2 golden agoutis. 2 yellow agoutis, 2 black-yellows and 2 albinos. The yellow-agoutis and black-yellows were clearly of the kind due to factor *f* and not to c^k or c^d. It was thus clear that ♀20213 was of constitution $Cc^a Ff$. The mutant spot can be explained as due either to a mutation from *F* to *f* in the ancestral cell, or to loss of the whole chromosome carrying *F*.

The case of ♂23538 is very similar to that just described. This animal, born April 20, 1924, in the control stock *B*, was originally called a golden-agouti tricolor. When three months old a conspicuous cream-agouti spot was observed on the left side (figure 6). As in the previous case, the appearance suggested factor *f*. Guinea pigs of formula *ff* typically show yellow of grades 6 to 8 at birth, but in a few cases there has been very little

dilution (grade 9) at this time. This probably accounts for the failure to note the spot earlier. Whatever the color at birth, there always seems to be a fading to pale cream, if not to white, later in life.

The parents were both golden-agouti tricolors. They produced 19 golden agoutis (including ♂23538) and 5 yellow agoutis, proving that both carried a dilution factor. The dam was a granddaughter of an *ff* dilute from mating *B*211, previously spoken of as the source of this factor. The

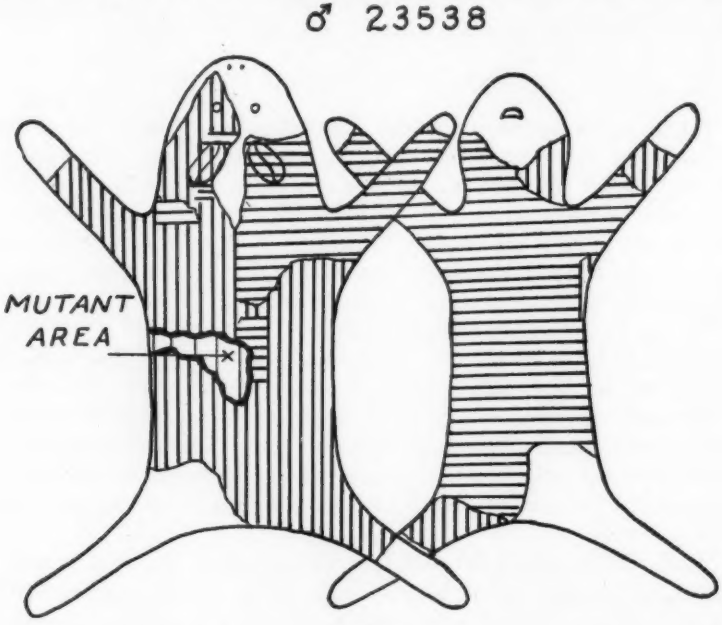


FIGURE 6.
The coat pattern of ♂ 23538, showing golden agouti (heavy vertical lines), cream agouti (light vertical lines) and red (horizontal lines).

sire traced through three generations to the mating which produced the female in *B*211 and which must thus have transmitted factor *f*. Dilution of the albino series (c^k) was also present on the sire's side, but seems to have been absent from the dam's pedigree. The sire was probably Cc^kFf, the dam *CCFf*. Male 23538 so far has produced 4 intense young from a mating with a black-yellow (*ff*) 4 intense from a meeting with an albino (c^ac^aFF) aside from four intense from a mating with an intense female. In all probability, he is *Ff* although the breeding test has not yet proved it.

The appearance of the cream-agouti spot suggests, as in the preceding case, that either *F* mutated to *f* or that the whole chromosome which carries it, was lost in the ancestral cell.

AN INTENSE BROWN-PALE BROWN MOSAIC ($Pp+pp$)

The last case is that of ♀25593, born September 5, 1924. She shows four colors, a typical intense brown and a sharply contrasting pale brown ex-

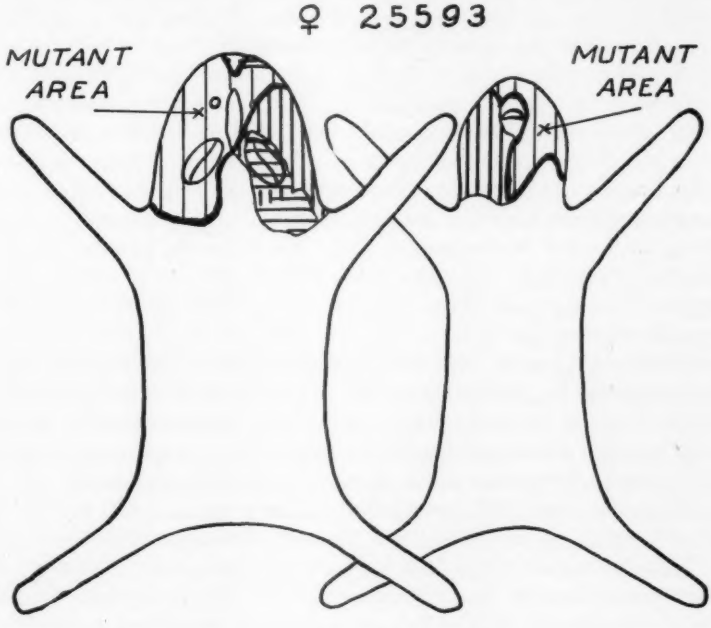


FIGURE 7.
The coat pattern of ♀ 25593 showing intense brown (heavy vertical lines), very pale brown (light vertical lines) and red (horizontal lines).

actly like that due to the pink eye factor *p*, in addition to the red and white of a tricolor (figure 7). The pale brown covers most of the head and left shoulder. An intense brown spot begins back of the right eye. Although the left eye is entirely surrounded by a pale brown fur and the right eye is more than half surrounded by it, the eyes are both of an intense brown, giving the head a very peculiar appearance to one familiar with guinea pig colors. The retina of the eye has, of course, such a different lineage,

ontogenetically, from the skin of the head that the lack of agreement requires no explanation except that required for the mosaic pattern as a whole.

This mosaic came from a back-cross involving six color factors, *SsEe^{p} AaCc^{k}BbPp*×*sse^{p}e$_{p}$aac^{k}c^{r}bbpp*. The appearance of ♀25593 indicates the constitution *sse^{p}e^{p}aa Cc^{kr}bb* (*Pp*+*pp*). The pale brown spots can be explained as due either to a mutation from *P* to *p* or to loss of the chromosome carrying *P*. In a mating with a pink-eyed dilute brown tricolor (*sse^{p}e^{p}aac^{k}c^{k}bbpp*) she has produced 6 brown-eyed young and 2 pink-eyed young, demonstrating that she transmits factor *P*.

DISCUSSION

Bud mutations and many cases of variegation have been reported in plants in which the abnormal parts have been shown to transmit a corresponding mutant character. These results indicate that mutation may take place at any time in the life history. In animals, we naturally look to Drosophila for the most extensive data. Here also many somatic mutations have been found (BRIDGES 1919; MULLER 1920). BRIDGES finds, however, that most mutations occur as single individuals which he interprets as meaning that mutation is most likely to occur at or very near maturation. He reports a few cases in which a fly has produced a number of mutant offspring, indicating that mutation relatively early in the germ tract had caused the individual to be germinal mosaic. He also reports mutants which appear as somatic mosaics due, presumably, to mutation subsequent to cleavage in a cell ancestral to the germ tract and part of the soma. MULLER (1920) reports similar cases as well as a case of a purely somatic mosaic. MOHR (1923) and MORGAN, BRIDGES and STURTEVANT (1925) report cases in which both soma and germ-plasm were mosaic, as in the mutant guinea pig No. 8400 reported here. MULLER points out that even if mutation is equally likely to occur at all stages of development, most mutations will actually be observed as single individuals, thus taking issue with BRIDGES' interpretation. Gynandromorphs in Drosophila are somatic mosaics which seem ordinarily to be due to dropping out of one of the *X* chromosomes in cells subsequent to cleavage (MORGAN and BRIDGES 1919). BREITENBECHER has reported numerous dominant purely somatic mutations in the cowpea weevil, Bruchus.

In vertebrates, relatively few such cases have been reported where there was any knowledge of the genetic situation. Three cases described by CASTLE and by CASTLE and PHILLIPS may be mentioned. One of these (CASTLE 1912) concerned a guinea pig which was close to an albino in

appearance but had small areas of sepia on the right side of the head, the hips and dark streaks in the iris of each eye. The parents were both colored but one at least was known to transmit albinism. CASTLE (1922) interprets the case as probably a heterozygote, $c^d c^a$, in which factor c^d was lost from a cell at a very early stage by non-disjunction. He explains similarly a tricolor rat, gray, yellow and white, known to be heterozygous for pink-eyed yellow (Pp). This rat had many separate yellow spots. Breeding tests carried to F_2 gave no indication that he transmitted any new factors. The third case occurred in CASTLE and PHILLIPS' experiments on changing the hooded pattern of rats by selection. In the tenth generation of plus selection an ordinary "plus" rat sired two mutants from different females and presumably many normal plus young. These mutants, intermediate between the plus hooded stock and self color, proved to be heterozygous for a new third allelomorph of the hooding factor, dominant over the latter but recessive to self color. This dominant mutation occurred apparently in at least two germ cells of the sire who was thus a germinal mosaic.

Returning to the guinea pigs described in the present paper no one explanation seems to fit the 7 mosaics discussed above. In the last three cases the mutant animals were certainly or very probably heterozygous in the factor affected (Ff two cases, Pp one case). The loss of the chromosome carrying the dominant factor in one of the somatic divisions seems to be the most plausible explanation, with mutation to the recessive as a possible alternative. The case of the black spot on a homozygous agouti (AA) requires the loss of two chromosomes (not necessarily simultaneously) or a double mutation or a mutation to an unknown dominant black. The two black-sepia mosaics were similarly homozygous in all known factors affecting intensity of color, but the possibility that the sepia was merely an aberrant effect of the tortoiseshell factor e^p, in which they were homozygous, makes them of little value.

The case of ♂8400 is the most remarkable one observed. His record may be summarized as follows:

(1) He was a dilute-intense mosaic somatically.

(2) He was produced by extensively tested dilute (recessive) parents each of which, however, had one intense parent.

(3) He transmitted the dominant mutant factor for intensity.

(4) He was a mosaic germinally. His germ cells were about one-third C, two-thirds c^d as indicated by a ratio of 79:149 in backcrosses.

(5) The proportion of C gametes seems to have varied from time to time.

(6) Both intense and dilute progeny bred normally.

These results can all be explained by the hypothesis that he was $c^d c^d$ initially but that one c^d mutated to C in a cell ancestral to part of the soma and also to part of the germinal epithelium and that the activity of different parts of the latter varied from time to time. The possibility of a rather complicated explanation involving aberrant chromosome behavior is not perhaps entirely ruled out.

The appearance of these mosaics throws some light on a number of questions. Factors which affect coat color may be conceived either as producing their effects by local action or by means of substances carried in the blood. The differently colored spots of a tricolor are quite obviously determined locally. Even here, however, it is not necessary to suppose that the piebald and tortoiseshell factors act locally. Sex is a factor which modifies the extent of colored spots in piebalds and of black spots in tortoiseshells but which need not be thought of as acting locally. The relatively uniform color differences due to factors *a*, c^d, *f*, *b*, and *p*, it would seem, might well be constitutional rather than local. Nevertheless, the cases of persistent somatic mosaics, in all probability due to mutation or loss of certain of these factors in particular somatic cells, indicate that the effects are produced locally or, in other words, that the pigment produced by a particular cell depends, in the main, on the genes in its own nucleus. The evidence is reasonably satisfactory for factors c^a, *f* and *p*, less so for *a*. No case apparently involving *b* has yet been found.

Another point of interest is the shape of the mutant spots. If each is due to a single mutation or chromosome loss, it appears that a single cell may give rise to rather widely scattered descendants. This seems to have been the case with CASTLE'S tricolor rat mosaic. The mutant areas of ♂8400 are widely scattered, as may be seen from the figure. The wide separation of the head and shoulder spots of red-agouti on ♂8400 from the rump spot is especially important in view of the completeness of this record, including evidence that his germinal epithelium was also mosaic.

The cream-agouti spots in ♀20213 and ♂23538 are very irregular in shape. In ♀20213 the cream-agouti is all on the left side of the head, but is broken into at least three portions, separated by red-agouti. In ♂ 23538, the spot runs irregularly from the mid-dorsal line to the belly down the middle of the side. In both of these cases, the shape is more irregular than is characteristic of either the colored or white spots of a piebald. Similar spots could perhaps be found on a tortoiseshell. The pale brown area of ♀25593 is compact enough except that it is cut by the white nose streak. The genetic condition of the mutant area doubtless is present

in this streak. Similarly, ♀2823 had two separate sepia spots but as these were separated merely by white, there is not necessarily any genetic discontinuity. Male 8210 and ♀2772 each had merely one compact mutant area.

Another curious fact is the relationship between certain of the mutants. Female 2823 (black-sepia) was the granddam of ♂8210, also black-sepia. Female 2772 agouti-black ($AA+aa?$) was the great-granddam of ♀20213 (red-cream-agouti) ($Ff+ff$) agouti and ♂23538 ($Ff+ff$) was related fairly closely to both of these. The case of the two black-sepias may, as noted in the text, involve merely inheritance of certain peculiar modifiers and not belong in the class of mosaics discussed here. The other cases may indicate an hereditary tendency toward germinal instability. LITTLE (1916) notes as possibly due to such a cause the occurrence of mutation from wild gray to brown-agouti (B to b) and from ordinary wild gray to white-bellied gray (A to A^1) in the descendants of a certain wild-gray mouse. The relationships here noted are rather closer than would be expected by chance in the large stock of guinea pigs of many distinct strains maintained by the BUREAU of ANIMAL INDUSTRY.

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THE GENETICS OF *LYCOPERSICUM ESCULENTUM* MILL
1. THE TRISOMIC INHERITANCE OF "DWARF"[1]

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Received November 23, 1925

The occurrence of triploidy in the tomato has recently been reported (LESLEY 1925). In 1923 one of two triploid plants of Dwarf Aristocrat, a dwarf variety with scarlet fruit, was crossed with Globe, a diploid with standard habit and crimson fruit. The dwarf habit is a simple recessive to standard, and the yellow skin of scarlet fruit is a simple dominant to the colorless skin of crimson. An F_1 of three plants was grown, all of which were different in appearance from the F_1 diploid Dwarf Aristocrat × Globe and also from one another. Two of them were found to have 26 chromosomes or 2 more than the diploid (somatic) number; as was to be expected from their origin, these plants were double trisomics. The third plant developed two cotyledons and one pair of first leaves but no bud. Its chromosome number was not determined.

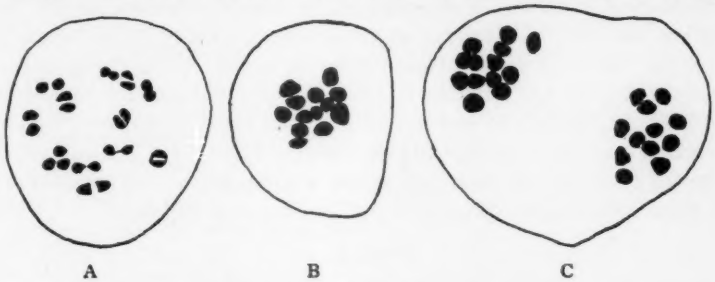
The more vigorous of the 26-chromosome plants, no. 23. 123.1, was a standard with scarlet fruit. It was less hairy than the diploid F_1, the leaves were decidedly thicker and more leathery in texture and their surface more rugose. The margin of the leaflets had fewer deep indentations and was more undulate. The lateral leaflets were narrower than in the diploid, but the terminal one was broader than the laterals and often somewhat twisted. The sepals were shorter and the petals somewhat deeper yellow, narrower at the base and blunter. The stamens were more slender, paler yellow and often incompletely syngynesious. The pollen was less in amount and contained more empty grains. The stigma, unlike that of the diploid, was usually below the apex of the staminal cone. One flower was entirely without a gynoecium. Under field conditions the plant was unfruitful unless artificially pollinated. The fruit was somewhat less regular in shape than that of the diploid F_1, and the seeds were relatively few.

This plant gave rise to an F_2 consisting of 41 plants, of which 3 were found to be simple trisomic, while the remainder were probably diploid. The diploids consisted of 22 standards and 16 dwarfs, a departure from

[1] Paper No. 140, UNIVERSITY of CALIFORNIA, Graduate School of Tropical Agriculture and CITRUS EXPERIMENT STATION, Riverside, California.

the disomic 3:1 ratio towards the trisomic ratio of 5:4 which would be expected to result if one of the two extra chromosomes of the parent were that containing the locus of the dwarf-standard pair of genes. Since the triploid P_1 was dwarf, the F_1 would contain two dwarf and one standard-bearing chromosomes or the genes *Ddd*. With regard to fruit color 27 plants were scarlet and 9 crimson, which corresponds exactly to the disomic ratio of 3:1.

One of the three simple trisomic plants, no. 24.094.32, was a standard. The figure was drawn with a camera lucida from smears made by Belling's



Chromosomes in pollen-mother-cells of simple trisomic tomato, 24.094.32. Stained in acetocarmine.

A. Diakinesis. Eleven bivalents and one trivalent. ×2450.

B. Metaphase of heterotype division. Twelve bivalents and one univalent at periphery and less deeply stained. ×3050.

C. Metaphase of homotype division. Extra chromosome present in one plate undivided. ×4000

iron-acetocarmine method. Although this plant was nearer its diploid sibs in hairiness and in color of corolla, it was decidedly slower-growing and later in flowering and in general much more nearly resembled its 26–chromosome parent. The foliage was similar in shape and texture, the sepals short and the stigma retruded. The style and stigma were perhaps more often misshapen than in the 26-chromosome F_1 plant. There was a similar tendency to pollen sterility and it set no fruit in the field without artificial pollination. The ripe fruit held tenaciously at the articulation. Apparently one of the extra chromosomes present in the 26-chromosome combination of plant 23.123.1 produced comparatively little external effect. It should be noted that both the F_1 plant 23.123.1 and the F_2 plant 24.094.32 although standard in general habit, showed certain dwarf-like characters, especially in the texture and rugosity of the leaves and in the short sepals and style.

In the selfed family from the simple trisomic 24.094.32, the germination was 94 percent and nearly all the seedlings survived. Among the diploid

progeny there were 17 standard and 20 dwarf plants, which again accords better with the 5:4 ratio expected in trisomic inheritance than with the disomic ratio of 3:1. The simple trisomic plants, expecially the standards, were clearly recognizable by their close resemblance to the parent 24.094. 32, and 6 of them were confirmed as such by chromosome counts. They consisted of 8 standard and 3 dwarf plants, a ratio which approaches the expected trisomic ratio of 7:2.

In a cross between Dwarf Champion and the same simple trisomic plant 24.094.32, the expected ratio among diploids was 1 standard to 2 dwarfs. The observed result was 14 standard and 29 dwarfs. The germination was excellent and every seedling survived but there were no trisomic progeny. A similar tendency to non-transmission of certain extra chromosomes through the pollen was found by BLAKESLEE (1921) in Datura, by DEVRIES and BOEDIJN (1923) in Oenothera, by CLAUSEN and GOODSPEED (1924) in Nicotiana and by FROST and MANN (1924) in Matthiola. Assuming this to be the case here, the data from selfing would indicate a transmission of the extra chromosome through about 23 percent of the eggs.

SUMMARY

Two 26-chromosome plants have been found in the F_1 progeny of triploid Dwarf Aristocrat × Globe. Selfing of one of these produced a simple trisomic plant of the standard type which closely resembled its double trisomic parent and differed in several respects from related diploids. It was slower-growing and showed certain dwarf-like characters. Breeding results show that the extra chromosome present in the simple trisomic was that containing the locus of the standard-dwarf pair of genes. The extra chromosome was not transmitted through the pollen but was present in about 23 percent of the progeny from selfing.

The writer gratefully acknowledges the help given by his wife, MARGARET MANN LESLEY, and by Professor R. E. CLAUSEN of the Division of Genetics, UNIVERSITY of CALIFORNIA, where part of this work was done.

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INHERITANCE OF PLUMAGE COLOR IN THE RHODE ISLAND RED BREED OF DOMESTIC FOWL[1,2]

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Received December 5, 1925

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INTRODUCTION

The color pattern in the Rhode Island Red is known to be highly variable. In an ordinary flock coming from "standard" ancestry may be seen birds of such a pale shade of red as to be almost buff, birds of about the color tone of English red, mahogany red birds, bay birds, etc. Auburn appears to be a rather common shade of color in the flock studied.

The amount of melanic pigment present fluctuates widely as shown by varying degrees of stippling, mottling, and ticking as well as by the occurrence of red feathers spangled with black. The variability in color pattern for this particular breed of fowl suggests that the so-called "standard" or ideal color pattern is of rather complex nature.

"Standard" color according to the 1923 edition of the Standard of Perfection is rich brilliant red and rich even red, except for the presence of black in the flight feathers and part of the flight coverts and tail in both sexes and in the lower neck feathers of the female. The shade of red preferred depends somewhat upon individual opinion, but at any

[1] Contribution No. 43 from the MASSACHUSETTS AGRICULTURAL EXPERIMENT STATION

[2] Prepared for the Kansas City meeting of the American Association for the Advancement of Science, December, 1925.

rate, bay is generally in greatest favor. The ideal specimen is apparently one showing the same shade of red over all sections of the body. In the study to be presented below, "standard" color is applied only to those birds closely approaching bay, in the surface plumage and carrying black in flight feathers and tail of both sexes and black in neck feathers of the female. The under-color will be specified as black or blue, smutty or light, if not red.

BITTENBENDER (1922) believes that a bar of slate or black pigment in the back feathers of either males or females used as breeders is necessary to secure rich, red plumage color. He states that only one parent should carry this pigment bar in mating for "standard" color. He emphasizes the importance of using both male and female breeders of the same color shade.

GOODALE (1910) has shown that the Brown Leghorn color pattern is sex-linked. He (1911) also refers to some F_2 birds in a cross between Brown Leghorn and Buff Plymouth Rocks as showing the Rhode Island Red color. Further details concerning this particular color have not been published.

AGAR (1924) makes reference to a pigment called "chestnut" which he discovered in crosses between Barred Rocks and Rhode Island Reds. This pigment is brown in color and is independent of the gene for gold (*s*) and apparently not sex-linked in its inheritance.

LLOYD-JONES (1915) found all pigment granules in red pigeons' feathers to be chestnut in color and globular in shape. His observations with those of LADEBECK (1922) would suggest that red in the pigeon and in the Rhode Island Red are similar in both morphology and color tone of pigment granules.

According to LADEBECK (1922), all red feathers from Rhode Island Reds contain reddish-brown to reddish-yellow pigment globules of melanic nature. The black feathers carry black pigment granules, some rod-shaped and some eliptical. In addition, the dark feathers carry pigment globules of varying sizes. The presence of both rod-shaped and eliptical pigment granules in the dark feathers appears to be characteristic of the Rhode Island Red.

DUNN (1922) has suggested that Rhode Island Reds carry the recessive gene e^m which restricts melanic pigment to the neck, wings, and tail. The normal allelomorph E^m is therefore not present in Rhode Island Reds so that the xanthic factor for red is not obscured. This variety therefore carries a definite pattern factor which restricts black pigment to neck, wings, and tail as called for in the standard color.

LIPPINCOTT (1923) has demonstrated that Rhode Island Reds carry a gene *E* which extends black pigment throughout the plumage. When genes E^m and *E* are brought together in the same individual, blue plumage results. In order to account for blue rather than black in such cases, it is necessary to assume that some restriction factor such as Lippincott's *R* is present so that the melanic pigment is so distributed as to give blue rather than black feathers.

STOCK USED IN THIS EXPERIMENT

The birds used in this experiment consisted of recessive whites and pure-bred Rhode Island Reds. The recessive white stock came from a White Plymouth Rock foundation crossed on Brown Leghorns and later bred back to Brown Leghorns and finally to Rhode Island Reds from which extracted whites came in later generations. Part of the foundation stock was developed by Dr. H. D. GOODALE. Breeding tests showed that these whites were free from such cryptomeres as barring, spangling, or silver. Such whites always bred as true recessives.

The Rhode Island Reds used consisted of two general classes, namely, a group with wide variability in color pattern that came from a "standard" foundation, that had been bred for fecundity for a period of ten years without attention to color pattern and two "standard" color males purchased from a prominent breeder. All Rhode Island Reds were pure-bred in the ordinary use of the term.

METHODS OF EXPERIMENTATION

In order to determine the genetic constitution of the Rhode Island Red for color pattern, it was necessary to cross different phenotypes on recessive whites and later to ascertain the genetic constitution of phenotypes resulting. It will be observed that Rhode Island Reds, crossed on recessive whites, give progeny that may be grouped into several general phenotypes. The plan was to test these different phenotypes by mating *inter se*. The experimental crossing began in 1923 and was continued in 1924 and 1925. All resulting progeny were described in adult plumage or at about 150 days old. Feather samples were taken at this time from five body regions. Feather samples were plucked from hackle, mid-back, main tail, breast, and body fluff. No attempt was made to record the juvenile plumage. All birds were pedigree hatched and were given a permanent wing and a permanent leg band each bearing the same number.

EXPERIMENTAL RESULTS

Proposed Theory

A study of the color pattern in progeny resulting from crosses made on three successive years suggests the following concerning the factor complex in color in Rhode Island Reds:

Gene *B* is an autosomal gene for reddish-brown pigment which distinguishes Rhode Island Reds from the gold of the Brown Leghorn or the buff found in the Orpington, Buff Leghorn, etc. Buffs have been shown by DUNN (1922) to be $e^m e^m ss$ but the relation between buff and gold has not been determined so far as I am aware. Gene *B* with either *L* or *E* or both always results in some shade of red on the surface. If present with factor *L* only the red pigment extends to the under color as well. If *B* is present, with factor *E* only, there will be brown stippling in the females. *B* in the presence of both *L* and *E* gives smutty under-color but a red surface. Factor *B* alone gives white, the presence of *L* and *E* in the absence of *B* gives the Brown Leghorn or Jungle fowl pattern, and either *L* or *E* alone gives white.

Gene *L* is a sex-linked gene for color pattern and gold color. Its presence makes possible the appearance of distinct color areas on wings, breast, etc., as in the Jungle fowl when the necessary pigment and extension factors are present.

Gene *E* is an autosomal gene for the extension of melanic pigment throughout the feathers such as LIPPINCOTT has pointed out in his studies on blue color.

Gene *R*, LIPPINCOTT'S factor for restriction of pigment within the feather and for the shape of pigment granule, is probably present in those birds showing blue smut in under-color.

Gene e^m is also present as DUNN has previously pointed out.

Gene *C* of HADLEY (1913) is also present.

The chief concern in this report is with genes *B*, *L*, and *E* as these three or their allelomorphs in the various combinations give the distinct phenotypes that make up Rhode Island Red color pattern. Attention is called to the fact that this study deals with distinct color phenotypes and has not been carried far enough to discover possible variations in shade of red that might result from multiple factors. Further experimental tests now being carried on should further elucidate this question.

The different phenotypes studied in this experiment together with their probable genetic composition are listed below for each sex.

COLOR ANALYSIS-PHENOTYPES AND GENOTYPES

Males

Phenotypes	Genotypes
Standard Red—No Smut	*BBLLee*
	BBLlee
Standard Red—No Smut	*BBllEE*
	BBllEe
Standard Red—Smutty	*BBLLEE*
	BBLLEe
	BBLlEE
	BBLlEe
Light Red—No Smut	*BbLLee*
	BbLlee
Medium Red—No Smut—Stippled	*BbllEE*
	BbllEe
Medium Red—Smutty	*BbLLEE*
	BbLLEe
	BbLlEE
	BbLlEe
Leghorn Color—Smutty Black Breast	*bbLLEE*
	bbLLEe
	bbLlEE
	bbLlEe
White	*BBllee* piled
	Bbllee piled
	bbLLee
	bbLlee
	bbllEE
	bbllEe
	bbllee

Females

Phenotypes	Genotypes
Standard Red—No Smut	*BBLoee*
Standard Red—No Smut—Stippled	*BBloEE*
	BBloEe
Standard Red—Smutty	*BBLoEE*
	BBLoEe stippled
Light Red—No Smut	*BbLoee*
Medium Red—No Smut—Stippled	*BbloEE*
	BbloEe

Medium Red Smutty	*BbLoEE* *BbLoEe* stippled
Leghorn color—Smutty—Stippled Salmon Color	*bbLoEE* *bbLoEe*
White	*BBloee* piled *Bbloee* piled *bbLoee* *bbloEE* *bbloEe* *bbloee*

Experimental Matings—1923

A series of eight matings gave progeny in the spring of 1923 but unfortunately only two resulted in families of sufficient size for genetic study. The total number of progeny described was thirty-one. The probable genetic formulae of the parents with the actual and expected phenotypes of progeny are given below:

TABLE 1

Piled white male C 4421 (*bbLlee*)×*Red, stippled, smutty female B* 9864 (*BbLoEe*).

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	2	1	5	3.5
Non-smutty	0	1	1	1.75
White	2	2	1	1.75
Standard red	0	0	2	0
Medium or light red	2	2	4	5.25
White	2	2	1	1.75
Stippled	2	1.5		
Non-stippled	0	.5		
White	2	2		

TABLE 2

Piled white male C 4421×*White female C* 3369

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
White	4	4	2	2

Experimental Matings—1924

In the spring of 1924 the total number of matings giving progeny was nine, with 54 cockerels and pullets described. There were five matings that gave sufficient numbers of adult progeny for study. The probable

genetic formulae of the parents together with the phenotypes of their progeny and the expected ratios are given below:

TABLE 3

Standard smutty Rhode Island Red male C 75450 (*BBLLEe*)×*White female C* 3369 (*bbloEe*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	3	2.25	1	2.25
Non-smutty	0	.75	2	.75
White	0	0	0	0
Medium red	3	3	3	3
Light red	0	0	0	0
White	0	0	0	0
Stippled	2	1.50		
Non-stippled	1	1.50		
White	0	0		

TABLE 4

White male C 9551 (*bbLlee*)×*Dark Rhode Island Red female C* 4518 (*BbLoEE*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	3	2	1	3
Non-smutty	1	1	2	0
White	0	1	0	0
Medium red	2	2	3	1.5
Light red	2	1	0	1.5
White	0	1	0	0
Stippled	1	3		
Non-stippled	3	0		
White	0	1		

TABLE 5

White Male C 9551 (*bbLlee*)×*Very dark Rhode Island Red female C* 5701 (*BbLoEe*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	4	2.25	3	2.50
Non-Smutty	5	4.50	2	1.25
White	0	2.25	0	1.25
Medium red	9	6.75	3	2.50
Light Red	0	0	2	1.25
White	0	2.25	0	1.25
Stippled	7	4.50		
Non-stippled	2	2.25		
White	0	2.25		

TABLE 6

Medium red smutty male C 9066 (BbLLEe)×White female C 8737 (bbloee)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	3	3	2	1
Non-smutty	1	1.5	0	.5
White	2	1.5	0	.5
Medium red	3	3	2	1
Light red	1	1.5	0	.5
White	2	1.5	0	.5
Stippled	4	3		
Non-stippled	0	1.5		
White	2	1.5		

TABLE 7

Black body, smutty male C 8619 (bbLlEe)×White female C 8473 (bbloee)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	1	.75	0	.75
Non-smutty	0	0	0	0
White	2	2.25	3	2.25
Standard red	0	0	0	0
Light red	1	.75	0	.75
White	2	2.25	3	2.25
Stippled	1	.75		
Non-stippled	0	0		
White	2	2.25		

Experimental Matings—1925

In the spring of 1925, twenty matings were made in this experiment. Of these matings sixteen gave progeny in fair numbers and are reported below. The total number of progeny described from the twenty matings is 88. A description of the parentage is given for the matings reported together with the actual and expected ratios of progeny.

TABLE 8

Medium red slightly smutty male E 1125 (BbLlEe)×Light red, stippled blue smutty female E 131 (bbLoEe)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Expected	
Smutty	1	1.13	4	3.75
Non-smutty	1	.56	0	.63
White	1	1.31	1	.62

TABLE 8 (continued)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Standard red	0	0	0	0
Medium and light red	2	1.69	4	4.37
White	1	1.31	1	.63
Stippled	2	1.50		
Non-stippled	0	.19		
White	1	1.31		

TABLE 9

Male E 1125 (*BbLlEe*) × *Dark red, stippled, black smutty female E* 132 (*BbLoEe*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	0	1.88	2	2.25
Non-smutty	2	1.88	0	.55
White	3	1.24	1	.20
Standard red	2	1.09	1	.75
Medium red	0	2.66	1	2.05
White	3	1.25	1	.20
Stippled	2	2.81		
Non-stippled	0	.94		
White	3	1.25		

TABLE 10

Male E 1125 (*BbLlEe*) × *Medium red, black smutty female E* 134 (*BbLoEE*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	5	2.50	2	2
Non-smutty	0	1.87	0	0
White	0	.63	0	0
Standard red	3	1.25	2	.50
Medium red	2	3.12	0	1.50
White	0	.63	0	0
Stippled	3	3.12		
Non-stippled	2	1.25		
White	0	.63		

TABLE 11

Male E 1125 (*BbLlEe*) × *Standard red, smutty female E* 480 (*BbLoEe*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	1	.75		
Non-smutty	0	.75		
White	1	.50		
Standard red	1	.75		
Medium red	0	.75		
White	1	.50		
Stippled	1	1.12		
Non-stippled	0	.38		
White	1	.50		

TABLE 12

Male E 1125 (*BbLlEe*) × *Standard red, non-smutty female E* 481 (*BBLoee*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty			2	1.0
Non-smutty			0	1.0
White			0	0
Standard			2	1
Medium			0	1
White			0	0

TABLE 13

Male E 1125 (*BbLlEe*) × *Light red, light under-color non-smutty female E* 482 (*BbLoee*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	0	.26	1	2
Non-smutty	0	.37	1	1.50
White	1	.37	2	.50
Standard red	0	.19	2	1
Medium red	0	.44	0	2.50
White	1	.37	2	.50
Stippled	0	.44		
Non-stippled	0	.19		
White	1	.37		

TABLE 14

Standard color non-smutty male E 485 (*BbLlee*)×*Medium red stippled non-smutty female E* 135 (*BBLoEe*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	0	.25	1	.75
Non-smutty	1	.50	2	1.50
White	0	.25	0	.75
Standard red	1	.37	1	1.12
Medium or light red	0	.37	2	1.12
White	0	.26	0	.76
Stippled	1	.50		
Non-stippled	0	.25		
White	0	.25		

TABLE 15

Male E 485 (*BbLlee*)×*Medium red, stippled blue smutty female E* 136 (*BbLoEe*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	0	.75	2	2.25
Non-smutty	1	.75	0	.56
White	1	.50	1	.19
Standard red	0	.44	1	.75
Medium red	1	1.06	1	2.06
White	1	.50	1	.19
Stippled	1	1.13		
Non-stippled	0	.37		
White	1	.50		

TABLE 16

Male E 485 (*BbLlee*)×*Medium red slight blue smut non-stippled female E* 407 (*BbLoEe*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	1	.13	1	.88
Non-smutty	0	.37	0	.62
White	0	.50	1	.50
Standard red	0	.13	1	.50
Medium red	1	.37	0	1.00
White	0	.50	1	.50
Stippled	0	0		
Non-stippled	1	.37		
White	0	.63		

TABLE 17

White male E 352 (bbLlee) × Standard color Rhode Island Red female C 7805 (BbLoEE)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	5	3	4	4
Non-smutty	1	1.5	0	0
White	0	1.5	0	0
Medium red	2	1.5	2	2
Light red	4	3	2	2
White	0	1.5	0	0
Stippled	3	3		
Non-stippled	3	1.5		
White	0	1.5		

TABLE 18

Male E 352 (bbLlee) × Pale color Rhode Island Red female C 9440 (BbloEe)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	2	.5		
Non-smutty	0	.5		
White	0	1.0		
Standard red	0	0		
Light red	2	1		
White	0	1		
Stippled	2	1		
Non-stippled	0	0		
White	0	1		

TABLE 19

Male E 352 (bbLlee) × Pale colored Rhode Island Red female C 9483 (BbloEe)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	1	.25	1	.50
Non-smutty	0	.25	1	.50
White	0	.50	0	1.00
Standard red	0	0	0	0
Medium or light red	1	.50	2	1
White	0	.50	0	1
Stippled	1	.37		
Non-stippled	0	.12		
White	0	.51		

TABLE 20

Spangled smutty male C 8619 (*bbL1Ee*) × *Pure white female C* 8473 (*bbloee*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	1	1.25	2	1.75
Non-smutty	0	0	0	0
White	4	3.75	5	5.25
Standard red	0	0	0	0
Medium or light red	1	1.25	2	1.75
White	4	3.75	5	5.25
Stippled	1	1.25		
Non-stippled	0	0		
White	4	3.75		

TABLE 21

Male C 8619 (*bbL1Ee*) × *Salmon, blue smutty, stippled female E* 225 (*bbLoEe*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	1	.38	1	1.50
Non-smutty	0	0	0	0
White	0	.62	1	.50
Standard red	0	0	0	0
Light red	1	.38	1	1.50
White	0	.62	1	.50
Stippled	1	.12		
Non-stippled	0	.25		
White	0	.63		

TABLE 22

Male C 8619 (*bbL1Ee*) × *White hens E* 353 *and E* 801 (*bbloee*)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	0	.75	0	.25
Non-smutty	0	0	0	0
White	3	2.25	1	.75
Standard red	0	0	0	0
Light red	0	.75	0	.25
White	3	2.25	1	.75
Stippled	0	0		
Non-stippled	0	.75		
White	3	2.25		

TABLE 23

Male C 8619 (bbLlEe) × Female C 8473 (bbloee)

CHARACTER	PULLETS		COCKERELS	
	Actual	Expected	Actual	Expected
Smutty	2	2	2	2.5
Non-smutty	0	0	0	0
White	6	6	8	7.5
Standard red	0	0	0	0
Medium red	2	2	2	2.5
White	6	6	8	7.5
Stippled	2	2		
Non-stippled	0	0		
White	6	6		

DISCUSSION OF RESULTS

Breeding tests demonstrate conclusively that color pattern in Rhode Island Reds is rather complex genetically. These tests help to explain why there is such a diversity in color pattern even in those flocks bred for "standard" color. In recent years, the trend of color shade has been from mahogany red to bay or even brown in some cases. There has been considerable difficulty in classifying birds as to medium or "standard" color. Such a classification is at best an arbitrary one and such discrepancies as are shown in the ratios are to be expected. This demand for deeper pigmentation has caused breeders to select as breeding stock those birds with the most intense black pigment in wings and tail and dark brown in body color. The use of some breeding stock carrying smut in under-color of back has also been common. Birds exhibiting a medium shade of red in surface plumage will actually appear darker in general plumage color if they carry black stippling in some degree or if they carry dark smut in under-color. Birds carrying black pigment in any part of the plumage, except the wings and tail or the neck of females, are considered "off color" but probably more desirable than birds deficient in black, and showing a light shade of red. The actual demand is for a deeper intensification of brown pigment granules and not for more black in the plumage.

The interaction of genetic factors involved in so far as they affect the color pattern really governs progress. These studies based on 173 progeny have strongly suggested that the true "standard" color Rhode Island Red male represents but four genotypes, namely, *BBLLee*, *BBLlee*, *BBllEE*, and *BBllEe*. That the true "standard" color female represents but three genotypes, namely, *BBLoee*, *BBLoEE*, and *BBloEe*. The un-

desirable smutty color makes its appearance whenever genes *B*, *L*, and *E* come together in the same zygote. Also, when genes *L* and *E* are present in the same zygote, the smutty under-color appears. Smutty under-color does not appear when genes *B* and *E* are brought together because *E* acts only in the presence of the pattern factor *L*. Males of the genetic formula *bbLLEE* will be of the same color as the Brown Leghorn or the Jungle fowl. Females of the genetic formula *bbLoEE* will be salmon in the body because of the presence of gold and smut.

Stippling has been observed to be of two general kinds, black and brown. Black stippling occurs most frequently on wings and breast in both males and females. Brown stippling appears to be confined to the female sex only. It may be restricted to the back and wings or may extend all over the body. Black stippled birds are probably those carrying genes *B*, *L*, and *E* while brown stippled birds appear to carry only genes *L* and *E*. The male phenotype that corresponds to the brown stippled female is apparently the black-breasted, light, smutty bodied male similar in color pattern to the Brown Leghorn male.

White birds are of several genotypes. They may carry any one of the three genes *B*, *L*, and *E* or may lack all and be true recessives. Birds of the composition *BBllee* are white because factor *B* acts only in the presence of *L* or *E*, *bbLLee* birds are white because the extension factor *E* is wanting and *bbllEE* birds are white because there can be no extension of pigment without the pattern factor*L*. True recessives are *bbllee* and breed as such. Even though they carry *C*, a gene for pigment, they lack the extension factor *E* and the pattern factor*L* upon which black pigment is superimposed.

APPLICATION OF THEORY TO BREEDING OPERATIONS

Theoretically the most desirable kind of "standard" color Rhode Island Red is the one in which genes *B* and *L* are combined. The genotypes would then be *BBLLee*, *BBLlee* males and *BBLoee* females. In such birds there should be no smut and there should be a rich red color throughout the plumage. Gene *B* behaves as a cumulative gene so that birds must be homozygous for *B* in order to be deep red. Birds heterozygous for *B* are light in shade of red. Individuals carrying genes *B* and *E* are less desirable for two reasons; first, there will be stippling in plumage; second, smutty progeny result whenever factor *E* is brought in contact with factor *L* or with both *B* and *L*. Females of the composition *BBLoee* would not breed true for Red because half their female progeny would be white when mated to *BBLl* males. Such females could be recognized if mated to

males of known genetic composition. *BBLoee* females, if mated to a "standard" smutty male *BBLlEe*, would give 25% white female progeny. If the male were homozygous for *E* (*BBLlEE*) there would be no white progeny. As the breed has progressed toward darker color, it is probable that most females as well as males carry gene *E*. This will account for the absence of white progeny and for the frequent occurrence of smut in any "standard" flock.

The crucial test for the selection of the breeding male is largely his phenotype or actual color. If he is rich bay in color throughout his feathers and is absolutely free from smut or stippling in any part, he should be either a *BBLLee* or a *BBLlee* bird. If in the breeding test using only females with deep bay surface color gives some white progeny, the probabilities are that the male is heterozygous for *L*. If the breeding test gives some smutty or stippled progeny, the gene *E* must be present in the female breeders.

The crucial test in the selection of female breeders is the individual color pattern. Deep bay, non-stippled, non-smutty females with red under-color are of only one genotype, *BBLoee*. Such females will not give smutty or stippled progeny unless gene *E* is introduced by their mate. Females may be deep bay in surface plumage but stippled with brown or black because of gene *E* or they may be smutty below the surface because they carry genes *B*, *L*, and *E*. The progeny of these *E*-bearing females will exhibit either stippling in females or smut in both males and females.

In breeding for rich bay plumage color with the columbian pattern, (gene e^m restricting black pigment to wings, tail and hackle) it becomes apparent that what is most necessary is the intensifying of brown color by the proper combination of genes *B* and *L*. The latter gene in addition to affecting color distribution, seems to intensify the shade of red so that *BBLL* birds are very dark while *BBllEe* birds are only medium red. *BbLLee* birds are too light in shade of red since they carry gene *B* in heterozygous condition. Another problem is to eliminate the gene *E* so that stippling or mottling and smutty under-color may no longer occur.

CONCLUSIONS

1. Rhode Island Red color pattern is a complex of several factors. Three factors are chiefly concerned in the make-up of the different phenotypes.

2. Bay pigment depends on the presence of a gene *B*, which is autosomal and epistatic to gold (*s*).

3. The sex-linked pattern factor *L* carries gold and to some extent influences the distribution of pigment and its intensity.

4. The autosomal gene *E* may or may not be present and determines the presence of smutty color in feather fluff.

5. Gene *E* is not essential in breeding for "standard" color because genes *B* and *L* together give the ideal color pattern.

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THE EFFECT OF ENVIRONMENT ON VARIEGATION PATTERNS IN MAIZE PERICARP[1]

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STATEMENT OF THE PROBLEM

EMERSON (1914, 1917) was the first to call attention to the remarkable series of multiple allelomorphs concerned with the development and distribution of color in maize pericarp. These allelomorphs include genes for self red, self orange, white-capped red, basal red, and colorless pericarp, associated with either self red or white cobs, and at least three types of variegated pericarp associated with similarly variegated cobs. The principal difference between these genes was thought to lie in their relative frequencies of mutation. It was suggested that the several types of variegated pericarp differ in frequency of factorial change by virtue of the fact that the change begins earlier in some types than in others.

HAYES (1917) made a study of a coarse type of variegation in maize, known as mosaic pericarp, with results quite similar to those found by EMERSON in the calico type of variegation. HAYES intercrossed the various

[1] This investigation was made in the Department of Field Crops, MISSOURI AGRICULTURAL EXPERIMENT STATION. The experiments were conducted at Sacaton, Arizona, under the supervision of C. J. KING, and at Chula Vista, California, under the supervision of C. G. MARSHALL, through the courtesy of the Office of Crop Acclimatization of the BUREAU of PLANT INDUSTRY.

allelomorphic types and concluded that certain combinations produce germinal instability.

EYSTER (1924) was able to demonstrate the suggestion made by EMERSON that the large series of multiple allelomorphs is evidence that a fundamental gene concerned in the development of pericarp and cob colors and color pattern has at one time or another mutated in diverse ways. A single self orange ear, in the course of a few generations in pedigreed cultures, gave rise to a large series of multiple allelomorphs, including strains of self orange varying in color intensity from whitish to deep red, self red, white-capped red, colorless and a relatively large number of distinct types of variegation, ranging in color pattern from ears that are almost colorless to ears that approach the self red ears in appearance.

It has also been found (EYSTER 1925) that the fundamental gene for mosaic pericarp changes in such a way as to give rise to pericarp and cob colors and color patterns which are in every respect parallel to those which occur in the calico type of variegation.

The detailed studies of both the calico and the mosaic series of multiple allelomorphs led the writer (loc. cit.) to the following general conclusions: (1) Self orange color is the expression of a gene which includes in its structure gene elements for red pericarp associated with gene elements for colorless pericarp, and (2) a variegation is produced when the contrasting gene elements become segregated by the mechanism of mitosis in the course of pericarp development.

The idea that a variegation is the result of the mitotic segregation of elements within the gene suggested the question as to the possible effects of different environmental conditions on the process. Accordingly, as a preliminary experiment, plantings from each of a number of variegated and self orange ears were made in localities which differ strikingly in such environmental conditions as might be expected to affect growth and development, and the present paper is a report of the results that have been found.

MATERIALS AND METHODS

The materials used in this experiment were prepared by crossing the plants having the pericarp color or pattern to be studied with a strain having colorless pericarp but red cob, and then back-crossing such F_1 plants with a strain having colorless pericarp and colorless cob, according to the method described elsewhere (EYSTER, 1924). The kernels of the variegated ears prepared in this manner were of two sorts as regards their genetic constitution, as follows: $\frac{W\ R}{W\ W}$ and $\frac{V\ V}{W\ W}$. Accordingly, approxi-

mately one-half of the plants produced ears with colorless pericarp and red cob, while the remainder of the plants produced ears with variegated pericarp and cob or some other allelomorphic type due to a genetic change in the *V V* gene. Likewise the kernels of the orange ears were of two sorts as regards their genetic constitution as follows: $\frac{W\ R}{W\ W}$ and $\frac{O\ O}{W\ W}$. In this case also, approximately one-half of the plants produced ears with colorless pericarp and red cob, while the remainder of the plants produced ears with orange pericarp and cob or an allelomorphic type due to a genetic change in the *O O* gene. By this method it was possible to identify the gene under study, regardless of what allelomorphic changes it may have undergone. In each case the ears with colorless pericarp and red cob, of the constitution $\frac{W\ R}{W\ W}$, were discarded.

It should be kept in mind that a single gene is responsible for the development and distribution of pigment in both pericarp and cob. The genes which have been referred to above are the following:

Symbol	Expression in Pericarp	Cob
W R	Colorless	Red
W W	Colorless	Colorless
V V	Variegated	Variegated
O O	Orange	Orange

Eight variegated and two orange ears were used in the experiment. The kernels of each ear were removed from the cob separately and divided into three equal lots, one lot to be planted in each of the localities chosen for growing the plants.

LOCALITIES IN WHICH THE PLANTS WERE GROWN

One lot of the kernels from each ear was grown in each of the following localities: Columbia, Missouri; Chula Vista, California; and Sacaton, Arizona.

Columbia, Missouri was chosen, largely as a matter of convenience, as a location where the environmental factors might be considered best suited for the normal growth and development of maize. Due to unfavorable cultural conditions and unusual chinch-bug injury, very few ears were matured in this locality, though more than 2500 plants were grown. Such ears as were produced in each of the progenies were quite like those grown at Chula Vista, California.

The planting at Chula Vista, California was made early in June, and the matured ears were harvested OCTOBER 15, making a growing season of approximately four months. During this period the air temperature seldom exceeded 75 degrees *F* and the mean daily fluctuation was not much over 20 degrees *F*.[2] The soil temperature had a daily range of from 60 degrees *F* to 80 degrees *F* and was very uniform. The radiation is less than that of the corn belt region due to the high relative humidity.

The planting at Sacaton, Arizona was made July 11 and the ears were fully matured October 15, making a growing season of a little more than three months. The air temperature was quite variable, ranging from 60 degrees *F* at night to as much as 110 degrees *F* during the day. The soil temperature was more variable, changing with the irrigations. The atmosphere at Sacaton is typical southwestern desert air, with a low relative humidity and a high radiation.

The soils in which the plantings were made at Chula Vista and Sacaton are quite comparable, both being alkaline and somewhat impervious. Both crops were grown under irrigation. The water used in irrigation at Chula Vista was surface water stored during the winter from mountain streams, while the water used at Sacaton was taken from wells and was distinctly salty. Both plots were irrigated before water stress conditions developed so that soil moisture conditions can be considered as having been optimum.

The chief environmental difference between Chula Vista and Sacaton seems to have been the higher and more variable temperature and greater transpiration at Sacaton. As a consequence of this difference the season required to mature the pericarp at Sacaton was fully a month shorter than that required at Chula Vista.

THE EFFECT OF ENVIRONMENTAL DIFFERENCE ON THE VARIEGATION PATTERN

The variegated ears grown at Chula Vista, in a cool, moist climate, where the growing season was relatively long, developed a fairly coarse pattern of variegation in the form of splashes, bands, and larger segments of the surface of a kernel, while the remainder of the pericarp appeared perfectly colorless. Variegated ears of the same genetic constitution and taken from the same parent ear but grown at Sacaton, in a region with a hot, dry climate and a short growing season, developed a strikingly

[2] The writer is indebted to J. H. Kempton of the Office of Crop Acclimatization of the Bureau of Plant Industry for the detailed statements concerning the environmental conditions existing at Chula Vista, California, and at Sacaton, Arizona.

different variegation pattern. The variegation consists in a smaller number of red areas which cover one-fourth and larger parts of the surface of a kernel, but a larger number of smaller red areas in the form of splashes,

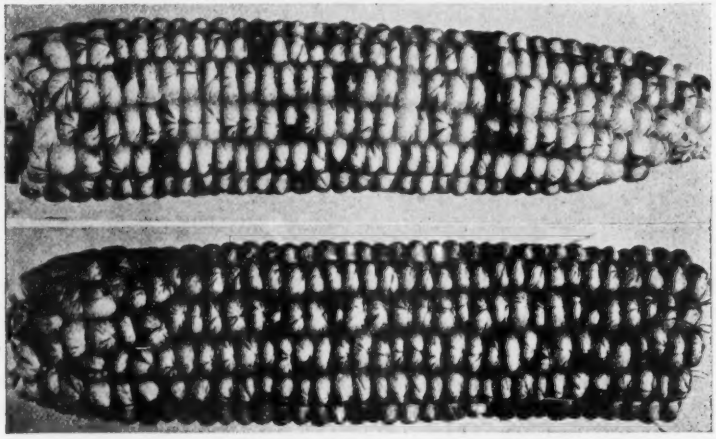

FIGURE 1.—Typical ears showing the variegation patterns of ears of the same strain grown under different environmental conditions. (*a*) Pattern which developed under Chula Vista conditions. (*b*) Pattern which developed under Sacaton conditions.

lines, and bands. The general surface of the kernels, not occupied by the red markings of the variegation is not colorless in the Arizona-grown ears as in the California-grown ears, but has a more or less distinct washed-out-

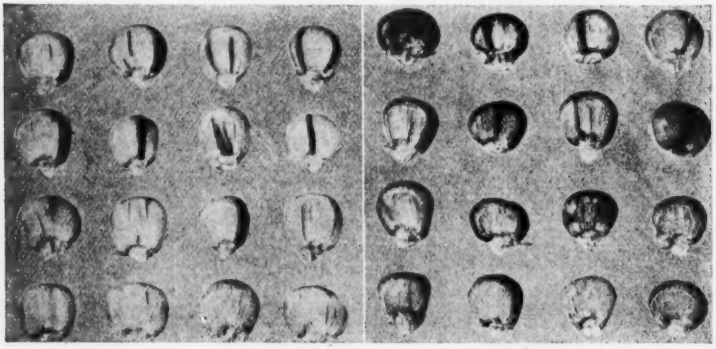

FIGURE 2.—(*a*) Representative kernels from ear shown in Fig. 1 a. (*b*) Representative kernels from ear shown in Fig. 1 b.

red color. Accordingly, all of the Arizona-grown variegations consist of relatively small areas of red on a washed-out-red background. The difference in the variegation pattern under the conditions of these two environ-

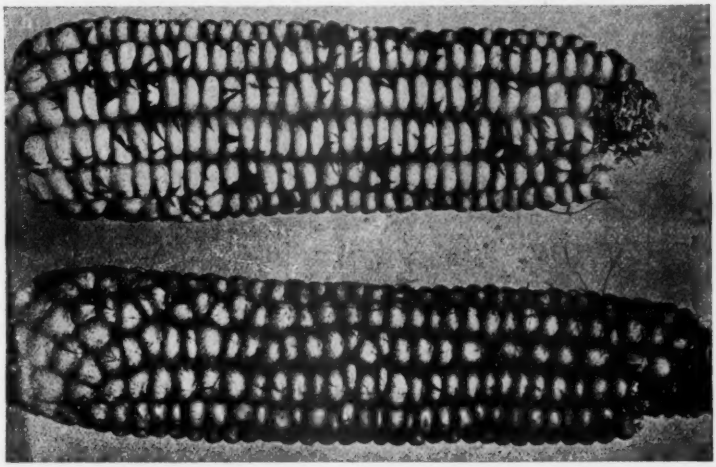

FIGURE 3.—Typical ears showing the variegation patterns of ears of a second strain grown under different environmental conditions. (*a*) Pattern which developed under Chula Vista conditions. (*b*) Pattern which developed under Sacaton conditions.

ments was consistent in all of the plantings that were made, as may be seen from the accompanying illustrations.

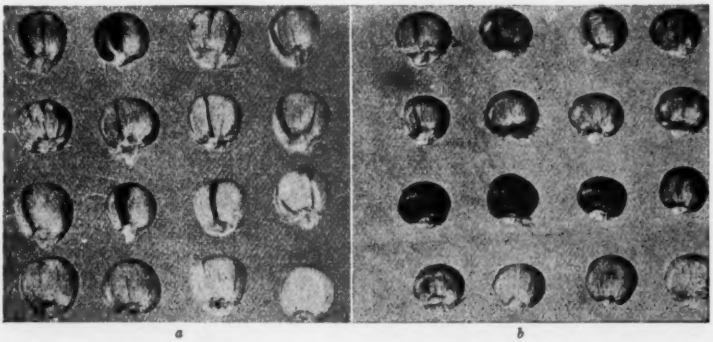

FIGURE 4.—(*a*) Representative kernels from ear shown in Fig. 3 *a*. (*b*) Representative kernels from ear shown in Fig. 3 *b*.

Kernels taken from the same parent ear were grown at Sacaton under number 2986 and at Chula Vista under number 2997. The ear shown in figure 1*a* illustrates the variegation pattern typical of the ears grown at Chula Vista, and ear figure 1*b* is representative of the ears grown at

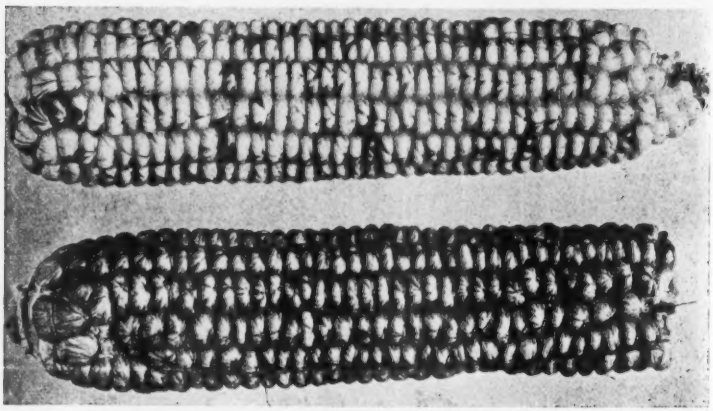

FIGURE 5.—Typical ears showing the variegation patterns of ears of a third strain grown under different environmental conditions. (*a*) Pattern which developed under Chula Vista conditions. (*b*) Pattern which developed under Sacaton conditions.

Sacaton. A comparison of typical kernels taken from these ears is shown in figure 2. The kernels grown at Chula Vista are shown in figure 2*a*, and those grown at Sacaton in figure 2*b*. The pattern differences which have already been described are apparent in these illustrations.

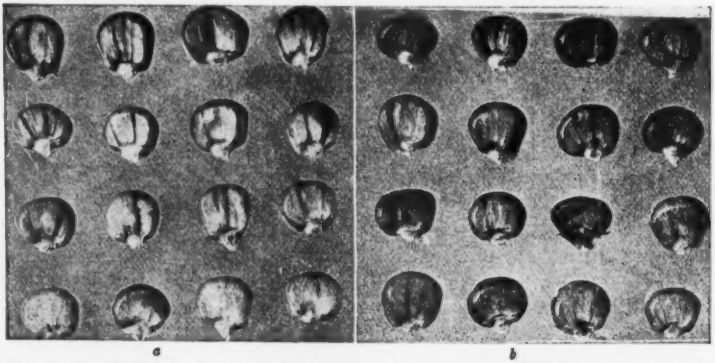

FIGURE 6.—(*a*) Representative kernels from ear shown in Fig. 5 *a*. (*b*) Representative kernels from ear shown in Fig. 5 *b*.

Kernels taken from a second variegated ear were grown at Chula Vista under number 2998 and at Sacaton under number 2987, and a typical ear from each planting is shown in figure 3. Typical kernels from these ears are illustrated in figure 4. The relatively large areas of red on a colorless background are characteristic features of the California-grown ears, and are in striking contrast with the fine markings and residual color in the pericarp of the Arizona-grown ears.

From a third variegated ear a planting was made at Chula Vista under number 2992 and at Sacaton under number 2982, and a representative ear from each planting is shown in figure 5. Representative kernels from each of these ears are illustrated in figure 6. Again the variegation patterns are strikingly different. Similar differences were found also in each of the other plantings that were made in connection with this experiment.

FREQUENCY OF OCCURRENCE OF SELF RED AREAS IN VARIEGATIONS UNDER THE CONDITIONS OF THE DIFFERENT LOCALITIES

The variegated ears were first studied for areas of red extending over more than a single kernel. The kernels were then shelled off the ear and studied individually and classified according to the size of the largest red area on each kernel.

California-grown Ears

From five to ten percent of the variegated ears grown in this locality had an exceedingly light pattern, while the remainder of the ears were like those shown in the illustrations above. The ears were separated into those which had the very light pattern and those which had the medium pattern like the parent ears before the data on the number and size of the red areas were taken.

A summary of the red areas found on the ears with the extremely light variegation pattern is given in table 1.

TABLE 1

PEDIGREE	VARIEGATED KERNELS	CHANGES TO RED IN KERNELS				
		¼	½	¾	1	>1
2997	2163	3	2	0	0	0
2998	1710	2	5	1	0	0
2990	2604	15	6	1	0	0
2992	203	7	3	1	1	0
2993	1126	3	4	2	1	0
2995	1178	3	2	0	0	0
Total	9584	33	22	5	2	0
percent	99.36	0.34	0.23	0.05	0.02	0.00

Two red areas were as large as a single kernel, but none were found that were larger than this. Less than one percent of the kernels had red areas that were as large as one-fourth of the surface of a single kernel.

The data on the number and size of the red areas that occurred on the more heavily variegated California-grown ears are given in table 2.

TABLE 2

PEDIGREE	VARIEGATED KERNELS	CHANGES TO RED IN KERNELS				
		¼	½	¾	1	>1
2997	8654	150	40	25	13	1
2998	6848	118	40	24	6	4
2990	10418	270	62	20	3	2
2992	5898	227	64	40	21	3
2993	4491	189	60	45	20	5
2995	6555	216	105	48	23	4
Total	42864	1170	371	202	86	19
percent	95.87	2.62	0.83	0.45	0.20	0.04

Nineteen red areas extended over more than a single kernel, and 4.13 percent of all the kernels had red areas that covered one-fourth the surface of a single kernel or larger areas.

Arizona-grown Ears

The variegated ears grown in Arizona did not include a single ear with the extremely light pattern found among the California-grown ears. The patterns were all fundamentally like those shown in the illustrations which have already been described, with variations in the number and size of the red areas and in the amount of residual color in the general pericarp.

A summary of the red areas that occurred on the variegated ears grown in Arizona is given in table 3.

Comparison of the Occurrence of Red Areas in California-and Arizona-grown Variegations

It should be kept in mind that the California-grown variegations were of two distinct patterns: (1) a medium heavy pattern, similar to that of the parent ear, and (2) an extremely light pattern. The variegations grown in Arizona were of one general type and consisted of many relatively fine red markings on a reddish background. A comparison in the frequency of the occurrence of red markings which covered one-fourth of a kernel and larger

areas in the heavy variegations grown in the two localities is shown in table 4.

In every planting that was made and studied there were more self red areas covering one-fourth of a kernel or larger areas in the California-grown variegations than in the corresponding variegations grown in Arizona. The differences range from 0.07 ± 0.17 to 2.56 ±0.27. In four of

TABLE 3

PEDIGREE	VARIEGATED KERNELS	CHANGES TO RED IN KERNELS				
		¼	½	¾	1	>1
2986	9254	96	21	16	18	1
2987	9624	154	51	39	19	0
2980	12211	143	60	22	5	1
2982	6215	107	58	19	16	0
2983	7123	246	105	41	37	4
2985	4500	92	43	16	13	0
Total	48927	838	338	153	108	6
Percent	97.14	1.66	0.67	0.30	0.22	0.01

TABLE 4

PEDIGREE.		CHANGES TO RED COVERING ¼ KERNEL AND LARGER AREAS, IN PERCENT.		DIFFERENCE
California	Arizona	California	Arizona	
2997	2986	2.58± .11	1.62± .09	0.96± .14
2998	2987	2.73± .13	2.66± .11	0.07± .17
2990	2980	3.31± .12	1.86± .08	1.45± .14
2992	2982	5.68± .20	3.12± .18	2.56± .27
2993	2983	6.63± .24	5.73± .18	0.90± .30
2995	2985	5.73± .19	3.52± .18	2.21± .26
Average		4.14±0.07	2.86±0.05	1.28± .086

the six plantings, the differences are from seven to a little more than ten times their probable errors and are doubtless significant. A total comparison of the six plantings shows that the percent of self red areas covering one-fourth or more of the surface of a kernel was 4.14 ± 0.07 under the California conditions and 2.86 ± 0.05 under the Arizona conditions. This is a difference of 1.28 ± 0.086. This difference is approximately 14.5 times as large as its probable error.

The objection might be made that by excluding the very light-patterned ears of the California-grown material an unfair comparison is made. Consequently in table 5 is given a summary of all the California-grown variegations, including the extremely light variegated ears, in comparison with the Arizona-grown variegations.

TABLE 5

PEDIGREE		CHANGES TO RED COVERING ¼ KERNEL AND LARGER AREAS, IN PERCENT.		DIFFERENCE
California	Arizona	California	Arizona	
2997	2986	2.12±0.09	1.62±0.09	0.50±0.13
2998	2987	2.30±0.11	2.66±0.11	−0.36±0.15
2990	2980	2.83±0.09	1.86±0.08	0.97±0.12
2992	2982	5.67±0.19	3.12±0.18	2.55±0.26
2993	2983	5.53±0.20	5.73±0.18	−0.20±0.27
2995	2985	4.59±0.15	3.52±0.18	1.07±0.23
Average		3.51±0.05	2.86±0.05	0.65±0.07

Again in four of the six plantings the California-grown kernels had a larger percent of kernels with red areas that covered one-fourth or more of the surface of a kernel. The differences in percent range from four to nearly ten times their probable errors, even when the extremely light variegated ears of the California-grown plants are included. The differences in families 2998–2987 and 2993–2983 are in favor of the Arizona-grown ears but are not significant.

When the extremely light and heavy variegations of the California-grown kernels are considered together, the self red areas amount to 3.51 ± 0.05 percent, as compared with 2.86 ± 0.05 percent in the Arizona-grown variegations. This is a difference of 0.65 ± 0.07 . This difference is a little more than nine times its probable error.

The result of this comparison is that the California-grown kernels have more red markings that cover one-fourth the surface of a kernel and larger areas than the Arizona-grown variegations.

VARIEGATED AREAS ON SELF ORANGE KERNELS

Orange kernels, of the genetic constitution that has already been indicated, were taken from the same ear and divided into three lots, one of which was planted in Missouri under number 2974, the second lot was planted in Arizona under number 2984, and the third lot was planted in California under number 2994. The number of ears that were produced

from the planting in Missouri was too small for further study. The ears of the Arizona and California plantings varied from whitish to deep orange or red, and the orange pericarp changed to variegation and self red in the manner described previously (EYSTER, 1925). No color changes were larger than the surface of a single kernel.

The kernels were removed from the ears and classified according to the presence and size of variegated areas as given in table 6.

TABLE 6

	CHANGES TO VARIEGATION.			TOTAL
	Orange kernels	Band to ¼ kernel	¼ to entire kernel	
California grown	14462	1492	153	16107
Arizona grown	14412	1252	90	15754

The relative frequencies of the changes from self orange to variegated pericarp in the California-and Arizona-grown ears may be seen more clearly from the comparison expressed in percent of the total number of kernels given in table 7.

TABLE 7

	PERCENT OF TOTAL KERNELS			TOTAL
	Orange kernels	Changes to Variegation		
		Band to ¼ kernel	¼ to 1 kernel	
California grown	89.79	9.26±0.16	0.95±0.05	10.21±0.16
Arizona grown	91.48	7.95±0.15	0.57±0.04	8.52±0.16
Difference	−1.69	1.31±0.22	0.38±0.06	1.69±0.23

The variegations which covered from a band to one-fourth of the surface of a kernel occurred 1.31 ± 0.22 percent more frequently in the pericarp of the California-grown ears. This difference is about six times its probable error. The variegations that extended over as much as one-fourth the surface of a kernel and larger areas could be recorded accurately, and it was found that the California-grown ears had 0.38 ± 0.06 percent more of these larger areas than the Arizona-grown ears. This difference is a little more than six times its probable error.

This comparison indicates that changes in pericarp color from orange to variegation occurred more frequently under the California conditions.

RED AREAS ON SELF ORANGE KERNELS

A record was also made of the color changes from orange to self red which covered a longitudinal band of the surface of a kernel and larger areas. No red area was found that extended over more than a single kernel. A summary of the red areas which occurred on orange pericarp is given in table 8.

TABLE 8

	CHANGES TO SELF RED			TOTAL
	Orange kernels	Band to ¼ kernel	¼ to entire kernel	
California grown	15059	934	114	16107
Arizona grown	15361	342	51	15754

A comparison of the frequency of occurrence of red areas in orange pericarp between the California-grown and Arizona-grown ears, expressed in percent of the total number of kernels observed, is given in table 9.

TABLE 9

	PERCENT OF TOTAL KERNELS			TOTAL
	Orange kernels	Changes to Self Red.		
		Band to ¼ kernel	¼ to 1 kernel	
California grown	93.56	5.80±0.12	0.71±0.04	6.51±0.13
Arizona grown	97.50	2.18±0.08	0.32±0.03	2.50±0.09
Difference	−3.94	3.62±0.14	0.39±0.05	4.01±0.16

The red areas which covered from a longitudinal band to one-fourth of the surface of a kernel occurred 3.62 = 0.14 percent more frequently in the pericarp of the California-grown ears. This difference is a little more than twenty-five times its probable error. The red areas which extended over areas larger than one-fourth of the surface of a single kernel occurred 0.39 = 0.05 percent more frequently under the California conditions. This difference is approximately eight times its probable error.

From these data it is to be concluded that the changes in pericarp color from orange to red occurred more frequently under the California conditions as compared with the conditions in Arizona.

DISCUSSION

In previous studies on variegation, especially those mentioned in an early part of this paper, results were reported which suggest that a variegation is caused by the segregation of contrasting substances in the same gene. The size of a color change is large or small depending upon whether the necessary segregation within the variegation gene occurs earlier or later in development. As was pointed out by EMERSON, the probability of pericarp color changes effecting the germ-plasm and thus becoming genetic increases as they extend over larger segments of the surface of a kernel. The higher percents of color changes extending over one-fourth of the surface of a kernel and larger areas in the variegated and self-orange strains grown in California, as compared with the same strains grown in Arizona, indicate that genetic changes occurred more frequently under the California conditions. Since the genetic constitution of the several strains grown in the two localities were identical, it must be concluded that the observed differences in the frequency of genetic changes were due to the differences in the environmental conditions of the two localities.

Although the Arizona-grown variegations have fewer red markings that extend over relatively large areas, they appear to have more total color than the California-grown variegations. This suggests that the minimum amount of the pigment-producing component of the variegation gene necessary to cause pigment to develop in the pericarp may vary under different environmental conditions. According to this view, some of the segments which become pigmented in Arizona remain colorless in California.

These preliminary studies indicate rather definitely that environmental factors not only may modify the expression of a character in the soma, but may also have a direct influence on the germ-plasm.

SUMMARY

1. Strains of maize with variegated pericarp and strains with orange pericarp were grown under strikingly different environmental conditions as a preliminary study of the relation of environmental factors on the somatic expression and genetic changes of these pericarp color patterns.

2. Each strain was grown at Chula Vista, California and at Sacaton, Arizona. The chief environmental difference between these two localities seems to have been the higher and more variable temperature and greater transpiration at Sacaton. As a consequence of this difference the season required to mature the pericarp at Sacaton was fully a month shorter than that required at Chula Vista.

3. Eight variegated strains were grown at each of these localities, and in every case there was a definite and consistent difference in the variegation pattern.

4. The California-grown variegations consistently had more red markings covering one-fourth the surface of a kernel and larger areas than the Arizona-grown variegations. The significance of this difference is that under the California conditions more of the color changes extended into the germ-plasm and thus became genetic.

5. Changes from orange to variegation, and from orange to red also occurred more frequently under the California conditions.

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LINKAGE STUDIES WITH THE TOMATO

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INTRODUCTION

Experimental genetic data have been collected systematically for several years from the writer's crosses of varieties of the garden tomato, *Lycopersicum esculentum*. These studies have involved many of the most common differential characters of the species, both qualitative and quantitative, commercial and otherwise. Of the more qualitative characters nine are sufficiently analysed to warrant a report of findings as to the manner of their inheritance. Other qualitative characters, some new mutants, and a number of more difficult size and shape complexes are being subjected to further analysis.

Genetics of the tomato, besides being of ultimate commercial importance, has an immediate and special scientific interest, since the species has many closely related species, an abundance of varieties, an exceptionally wide range of size and shape differences in fruits suitable for quantitative studies, rather frequent mutations both by seed and by bud, and a reasonably small chromosome number ($n=12$), and is frequently used in studies of selective fertilization, graft-hybridization (WINKLER, 1916), and triploidy and tetraploidy (most recently by LESLEY and MANN

1925, and Mrs. LESLEY 1925). It thus promises contributions toward the solution of many current genetic problems from nearly all angles of approach.

The present report is designed to help establish the fundamental genetic pattern of the species. Accordingly, first attention has been given to the question of the independence or association in inheritance of certain well-defined factors.

DESCRIPTION OF CHARACTERS AND VARIETIES USED

The manner of inheritance of many characters of the tomato has been determined in part, and often only in a preliminary way, by earlier investigators chiefly from the AGRICULTURAL EXPERIMENT STATIONS, that is (HALSTED 1905, 1918), HURST (1906), CRAIG (1907), HEDRICK and BOOTH (1907), PRICE and DRINKARD (1908), GILBERT (1912), GROTH (1910–1915), CRANE (1915), FRIMMEL (1922), LINDSTROM(1924–1925), and WARREN (1925). The meagre and more or less unsatisfactory data were summarized for indications of linkage up to 1917 by JONES and up to 1925 in LINDSTROM'S more critical paper.

The characters I am reporting here concern differences in stature, fruit colors, fruit skin surface, distinct fruit shapes, form of inflorescence, foliage color and shape of leaf. In the brief descriptions which follow of the 9 distinct contrasting allelomorphic pairs, the abbreviations designating factors are, whenever possible, in order to prevent unnecessary future confusion, taken from former published work.

1. Stature (Vine habit). Tall (*D*) dominant over dwarf (d). Dwarf plants differ from the tall in having an upright habit of growth, stocky shortened stems, short broad cotyledons, leaflets and petioles, peculiar rugose, puckered and down-curved leaves, and a deeper green foliage color.

2. Fruit Skin Structure. Smooth (*P*) is dominant over "peach" (p). The "peach" fruit epidermis possesses a persistent pubescence and a glandular structure making the fruit tomentose and dull as compared with the smooth and shining appearance of fruit of other varieties. The gene *p* also produces some extra pubescence and a blue-gray cast over the entire plant.

3. Fruit Shape. Mature fruit without constricted neck (P_r) is dominant over pear-shaped mature fruit (p_r). Some confusion has formerly resulted from attempts to classify plants for these characters before their fruits attain maturity. Fruits of oval or plum shape often pass through an unripe stage resembling the true pyriform. Certain evidence suggests also that related fruit forms, such as "plum" and the slender-necked "calabash", carry factors allelomorphic with P_r and p_r.

4. Inflorescence Type. Simple inflorescence, arising from successive third internodes (*S*) is dominant over the compound, repeatedly branching, more or less leafy type arising from successive sixth nodes (*s*). Such a compound form with 30 or more flowers and fruits per cluster is found in one American variety, Grape Cluster, and in a few European varieties (CRANE 1915).

5. Fruit Flesh Color. Red flesh (*R*) is dominant over yellow (*r*). In mature fruit cells the gene *R* controls the formation of a red pigment (*lycopersicin*) in needle-like crystals, while its absence, or the presence of *r*, results in yellow granules. In "albino" fruited varieties the flesh color may possibly be a slightly paler yellow.

6. Fruit Skin Color. Presence of a yellow-orange pigment in epidermal cell walls of ripe fruit (*Y*) is dominant over the clear, colorless state (*y*).

7. Foliage Color. The ordinary fully-green foliage (*G*) is dominant over the pale yellow-green condition (*g*) met with in Honor Bright, whose new growth, at first green, soon becomes yellowish from below upward. Immature fruits of yellow-green plants remain characteristically pale white until just before ripening to their definitive color.

8. Leaf Shape. The deeply dentate leaflet of common "cut leaf" varieties (*C*) is partially dominant over "potato-leaf" (*c*) whose leaflets have nearly entire margins.

9. Fruit Shape. Fruits with smooth regular outline (*F*) are dominant over those of Albino, a Ponderosa type, with distinctly lobulated irregular contour (*f*), the irregularity even extending to the extreme of producing compound fruits by fusion of as many as four. This roughness of outline is anticipated even in the flower by a strongly correlated flattening of the pistil, a high number of petals and sepals (8–10 or more instead of 5–7) and in the fruit by a very large number of ovarian locules. The genes *Ff* are probably one of the complementary pairs described by WARREN (1925) as controlling fasciation, but in the absence of published quantitative data in his paper it is not yet possible to homologize the genes definitely.

The ten varieties (Vaughan's, Ferry's and Steele-Briggs') used containing these characters were all bagged and selfed for 2–5 years before crossing, and all proved homozygous for the characters dealt with. The genetic formulae assigned to these varieties below describe them most briefly:

Variety and Abbreviation		Constitution								
Dwarf Aristocrat	(Dw Ar)	*DD*	*PP*	P_rP_r	*SS*	*RR*	*YY*	*GG*	*CC*	*FF*
Pink Peach	(P Pch)	*DD*	p_hp_h	P_rP_r	*SS*	*RR*	*yy*	*GG*	*CC*	
Yellow Pear	(Y Pr)	*DD*	*PP*	p_rp_r	*SS*	*rr*	*YY*	*GG*	*CC*	*FF*

Variety and Abbreviation		Constitution								
McMullens Pink	(McM P)	*DD*	*PP*	P_rP_r	*SS*	*RR*	*yy*	*GG*	*cc*	*FF*
Albino	(Alb)	*DD*	*PP*	P_rP_r	*SS*	*rr*	*yy*	*GG*	*CC*	*ff*
White Apple	(Wh App)	*DD*	*PP*	P_rP_r	*SS*	*rr*	*yy*	*GG*	*CC*	*FF*
Honor Bright	(H Br)	*DD*	*PP*	P_rP_r	*SS*	*RR*	*YY*	*gg*	*CC*	*FF*
Golden Queen	(G Q)	*DD*	*PP*	P_rP_r	*SS*	*rr*	*yy*	*GG*	*CC*	*FF*
Red Currant	(R C)	*DD*	*PP*	P_rP_r	*SS*	*RR*	*YY*	*GG*	*CC*	*FF*
Grape Cluster	(G Cl)	*DD*	*PP*	P_rP_r	*ss*	*RR*	*YY*	*GG*	*CC*	*FF*

Since any one commercial variety seldom contains many recessive factors it was necessary to make scores of crosses to obtain all the desired combinations. From F_2 recombinations and successive synthetic crosses many new poly-recessive types are now available, including linkage testers for back-crossing.

The Unifactorial Nature of these Characters

That each of the characters described above is controlled by a single factor pair is quite fairly demonstrated from much F_2 and back-cross data in the crosses involving these allelomorphs. Experience has shown that dominance is nearly complete in the first generations of all, that the parental phenotypes reappear well-defined in the F_2 generation giving a clear segregation on a monohybrid basis, and that extracted recessives breed true in progeny tests.

The segregation is apparent from observation alone, but in all cases where fruit shapes and sizes are concerned confirmatory check measurements of polar and equatorial diameters and average fruit weights of all plants in all generations were also taken. Such data will be reported in later quantitative studies.

It might be noted that the data summarized in table 1 are the same as those used in dihybrid form for linkage tests (page 391) and include every classified individual of the F_2 generations bred, there being practically no mortality after germination.

TABLE 1

Monohybrid ratios obtained in F_2 populations

CHARACTERS AND FACTORS		F_2 PHENOTYPES		DEVIATIONS FROM EXPECTED 3:1 RATIOS
		D	*R*	
Tall (*DD*)	× dwarf (*dd*)	937	336	17.75±10.6
Smooth (*PP*)	× peach (p_rp_r)	976	324	1.0 ±10.51
Not pear (P_rP_r)	× pear (p_rp_r)	959	326	4.75±10.5
Simple (*SS*)	× compound (*ss*)	84	24	4.0 ± 2.93
Yellow (*YY*)	× clear (*yy*)	2339	725	41.5 ±15.86
Red (*RR*)	× yellow (*rr*)	1737	590	8.2 ±14.15
Cut leaf (*CC*)	× potato (*cc*)	560	172	11.0 ± 7.82
Green (*GG*)	× yellow (*gg*)	873	286	3.75± 9.89
Regular (*FF*)	× fasciated (*ff*)	305	111	7.0 ± 6.08

Since the deviations from the theoretical 3:1 ratios seldom much exceed those to be expected from random sampling, there can be no reasonable doubt that we are dealing in all these cases with single factor differences. The rather large departure from expectation in the crosses of yellow-skinned (*YY*) with clear-skinned (*yy*) varieties may signify a slight inviability of the recessive type or a small amount of selective fertilization favoring the *Y* gene.

TESTS FOR LINKAGE OR INDEPENDENCE OF GENES

The dihybrid and polyhybrid crosses so far carried through the F_2 generation furnish linkage tests covering 24 different combinations of these genes taken two at a time. The F_2 classifications were made by the writer and independently checked by Mr. CECIL DUFF, who gave valuable assistance in the work for three successive summers.

Four of the combinations of pairs of genes gave clear indications of linkage, while the remaining twenty combinations afford equally clear evidence of free assortment. The cases showing linkage will be first described.

THE FIRST LINKAGE GROUP

The first linkage group in the tomato contains genes *Dd*, *P p*, *P$_r$p$_r$*, and *Ss*. In the four crosses made involving two pairs of these genes at a time, in each instance one pair of genes entered with one parent and the other with the second; accordingly, the resulting F_2 ratios fit into the "repulsion" series. The linkages are close enough to be indisputable even without more critical backcross data, for the percentage of crossing over in no case exceeds 20 percent.

1. Dwarf Aristocrat (*dd PP*) times Pink Peach (*DD pp*). From this cross and its reciprocal all F_1 plants (*Dd Pp*) were tall and bore smooth fruits. The F_2 generation produced only three of the expected four classes (table 2).

TABLE 2

Data showing repulsion between d and p genes

CROSSES	F_2 CLASSES (PHENOTYPES)			
	DP	*Dp*	*dP*	*dp*
dd PP × *DD pp*	135	52	53	0
DD pp × *dd PP*	116	58	54	0
Total	251	110	107	0

This ratio evidently corresponds to a 2:1:1:0 ratio, suggesting very close or complete linkage (repulsion) between genes *Dd* and *Pp*. Presumably owing to this fact there is no known "dwarf peach" commercial variety of the tomato. Whether the linkage is actually complete, larger F_2 generations should determine. To date only 677 F_2 individuals from this cross have been reported classified, 468 in the present case and 209 in a paper by LINDSTROM (1925). The latter were distributed in a similar ratio: 101 DP:56 Dp:52 dP:0 dp, again indicating complete linkage; but from one of his F_2 plants (open pollinated) there segregated two double recessive, dwarf peach plants!

2. Dwarf Aristocrat (ddP_rP_r) × Yellow Pear (DDp_rp_r). The F_1 plants (DdP_rp_r) were tall in stature and produced small, more or less elongated fruits without constricted necks. Their progeny fell into four distinct classes as regards these characters: 106 tall, not pear (DP_r). 58 tall, pear (Dp_r) 57 dwarf, not pear (dP_r), and 1 dwarf pear (dp_r). From the reciprocal cross it is known only that 2 dwarf pear plants appeared in a total of 250 F_2 individuals.

The ratio, 106:58:57:1, with a coefficient of association of .934 corresponds approximately to a repulsion ratio with 13 percent crossing over. If the frequency of the fourth class is used as a criterion there are in both crosses 3 double recessives in the total of 474 F_2 plants, suggesting about 16 percent crossing over. The corresponding F_2 ratio obtained by HEDRICK and BOOTH (1907) was: 252:127:70:3, indicating ($Q = .843$) about 20 percent crossing over (JONES, 1917). Forthcoming backcross generations should determine the precise crossover value in the range from 13 to 20 percent.

3. Pink Peach (ppP_rP_r) × Yellow Pear (PPp_rp_r). The F_1 plants, of the double dominant phenotype (PP_r), gave four classes among their 498 offspring (table 3).

TABLE 3

Data showing repulsion between p and p_r genes

CROSSES	F_1 PHENOTYPES			
	$P\,P_r$	$P\,p_r$	$p\,P_r$	$p\,p_r$
Pink Peach × Yellow Pear	145	74	72	3
Yellow Pear × Pink Peach	102	51	50	1
Total	247	125	122	4

This ratio with a coefficient of association of .878 indicates a cross-over value of 17+percent, thus approximating, as theoretical considerations

require, that for the previous cross. For, if peach and dwarf are completely linked, they should show equally close linkage with a third factor pair (P_r p_r).

4. Dwarf Aristocrat (*ddSS*) × Grape Cluster (*DDss*). F_1 plants from this cross were all tall with simple inflorescence, and a small F_2 generation (112 plants) segregated sharply into the expected classes: 59 tall with simple inflorescence: 23 tall with compound inflorescence: 29 dwarf simple: 1 dwarf compound. The ratio with $Q = .837$ fits into the repulsion series with 20 percent cross-overs.

The evidence presented from these four crosses would seem adequate to justify the establishment of a linkage group containing the factors: *Dd*, *Pp*, $P_r p_r$, and *Ss*. In all instances the linkage is close and the quantitative values within the group mutually consistent.

Crosses involving *Ss* with *Pp* and with $P_r p_r$ and back-crosses involving all four pairs of linked genes, excepting *Dd* and *Pp*, are now in culture for the exact determination of loci and mapping of the "chromosome."

Facts are yet lacking even on such essential points as whether the loci of *Ss* and $P_r p_r$ are on the same or opposite sides of the closely linked peach and dwarf genes, though CRANE's findings might barely suggest as a working hypothesis that *Ss* and $P_r p_r$ are not at all close together.[1]

Exception has been taken by LINDSTROM (1925) to admitting linkage data on the pyriform factor, p_r, since "the heredity of fruit shape is relatively complex and has not been analyzed into its component parts." Now fruit shape is undeniably a product of many coöperating factors, but it is surely justifiable to utilize genetic elements of this complex as soon as they can be identified and dissociated from the unresolved residue, as, for instance, the genes for pear shape (p_r), for irregular outline of fruit (*f*), and probably also the complementary pairs for depth (*Aa* and *Bb*), and for fasciation (*Ee* and *Ff*) described by WARREN (1925). In the first two of these cases a fairly unmistakable segregation occurs making practicable a correct unbiased classification, as we have repeatedly tested. Accordingly I have dealt with them as distinct entities.

As regards the reported complete linkage, in another group, of the factors (*Gg*) for foliage color and those assumed to control number of fruit locules (PRICE and DRINKARD, 1908, and JONES, 1917), I found that the F_2 green-foliaged and yellow-foliaged segregates have fruits with the same average locule number. Only one linkage group in the tomato may thereore, be considered established.

[1] Backcrosses of the past summer demonstrate crossover values as follows: between *d* and p_r, $12 \pm 1.5\%$; between *p* and p_r, $8 \pm 1.0\%$; and between *d* and *p* (calculated), $4 \pm 1.8\%$, a value to be compared favorably with the 2.5 % observed by LINDSTROM (1926, Res. Bull. No. 93, Iowa Agric. Exp. Sta., page 104). Hence the order of genes is *d*——*p*——p_r——*s* (?)

The Combinations Showing Independent Assortment

The remaining 5 pairs of factors under analysis have turned out to be inherited independently of the 4 linked genes of the first linkage group and also independently of each other in all of the 20 different combinations (not counting reciprocals) of the 9 pairs so far bred through the second generations (see table 4). These are treated as dihybrid crosses though many dihybrid ratios were extracted from trihybrid or even tetrahybrid distributions.

TABLE 4

Combinations whose F_2 ratios indicate absence (—) of detectable linkage between pairs of genes.

	Rr	*Yy*	*Gg*	*Cc*	*Ff*
Dd	—	—		—	—
Pp	—	—	—		
P_rP_r	—	—	—	—	
Rr		—		—	—
Yy				—	—
Gg	—?	—		—	—

Since different methods of calculating the goodness of fit of the observed ratios to the 9:3:3:1 ratios expected in F_2 generations produced by free assortment of F_1 genes, give somewhat different results, it has seemed best to calculate both deviations from the 10:6 ($AB+ab$:$Ab+aB$) ratio and values for X^2 and P by the PEARSON-ELDERTON method. The proofs of random dihybrid distribution afforded by the data are summarized in tables 5–10 and detailed in the descriptions of crosses which follow, and in tables 11–18 to the Appendix.

Genetic Relations between Genes *Dd* and *Rr*, *Yy*, *Cc* and *Ff*

The genetic relations existing between the factor for dwarfness and those controlling fruit flesh and skin color, leaf shape, and fruit outline are revealed in the data of table 5.

***Stature and Flesh Color*—*Dd* and *Rr* genes.** Dwarf Aristocrat, a dwarf, red-fleshed variety (*ddRR*), crossed with tall yellow-fleshed sorts (*DDrr*) like Yellow Pear and Albino, gave all tall and red-fleshed F_1 hybrids, and these in turn an apparently freely segregating F_2 generation, as detailed in table 11 and summarized along with the three following combinations in table 5. The deviation from the 10:6 ratio is less than twice

the probable error, while $X^2 = 3.734$ and $P = .297$, showing a fair probability that the 9:3:3:1 ratio correctly expresses the results observed.

Stature and Fruit Skin Color—Dd and *Yy* genes. Dwarf Aristocrat is yellow-skinned (*ddYY*), and its four separate crosses with tall clear-skinned varieties (*DDyy*) as Pink Peach, McMullen's Pink, and Albino, produced tall yellow-skinned hybrids, whose progeny were distributed in four classes with numerical frequencies fitting expectation excellently for X^2 is only 0.5134 and $P > .801$ (table 12).

Stature and Leaf Shape—Dd and *Cc* genes. Dwarf Aristocrat (*ddCC*) crossed with the tall "potato-leafed" McMULLEN's Pink (*DDcc*) resulted in tall cut-leafed hybrids whose offspring fell into four sharply defined phenotypes: 132 tall cut-leaf: 31 tall potato-leaf: 53 dwarf cut-leaf: 17 dwarf potato-leaf; these numbers accord well with the 10:6 ratio but fit only fairly well by the X^2 method. There is no sign of linkage by either method.

Stature and Fruit Contour—Dd and *Ff* genes. Dwarf Aristocrat is quite smooth fruited (*ddFF*) as compared with the Albino (*DDff*). Their tall hybrids are also relatively smooth fruited, and the two pairs of factors segregate entirely at random in the second generation, there being 119 tall smooth (*DF*): 44 tall irregular (*Df*):42 dwarf smooth (*dF*):14 dwarf irregular (*df*), an excellent fit of observation to theory.

TABLE 5

Summary of data from linkage tests between Dd, and Rr, Yy, Cc, and Ff genes.

FACTORS INVOLVED WITH *Dd*		F_2 CLASSES				TOTAL	DEVIATION FROM 10.6 RATIO	X^2	P
		DX	*Dx*	*dX*	*dx*				
Rr	observed	231	96	83	31	441	13.6± 7.0	3.734	.297
	calculated	248	82.7	82.7	27.6				
Yy	observed	519	164	175	55	913	3.4± 3.14	0.513	>.801
	calculated	513.6	171.2	171.2	57.0				
Cc	observed	132	31	53	17	233	3.4± 4.9	6.071	.110
	calculated	131.0	43.7	43.7	14.6				
Ff	observed	119	44	42	14	219	4.0± 4.9	0.460	>.801
	calculated	123.3	41	41	13.7				

[1] *Xx* stands for any factor pair in the first column.

Genetic Relations between Genes *Pp* and *Rr*, *Yy* and *Gg*

In the various "peach" crosses the factors for skin structure (pubescence) were brought into combination with those mentioned above producing contrasting fruit flesh and fruit skin colors and foliage colors. From these combinations the genes reassorted freely in each case (table 6 for summary).

Fruit Skin Pubescence and Flesh Color—Pp and *Rr* genes. The Pink Peach-Yellow Pear cross (page 392) also brought together genotypes, *ddRR* and *PPrr*, whose elements redistributed at random in an F_2 population numbering 498 plants. These contained 288 smooth red (*PR*):81 smooth yellow (*Pr*): 92 peach red (*pR*):37 peach yellow (*pr*), which ratio differs from the expected chiefly in a slight deficiency of the *r* classes.

Fruit Skin Pubescence and Skin Color—Pp and *Yy* genes. Pink Peach, a double recessive (*ppyy*), was crossed with several varieties bearing fruits with smooth yellow skins (*PPYY*), as Dwarf Aristocrat, Yellow Pear, and Honor Bright. Their hybrids bore smooth, yellow-skinned fruit, and the F_2 generation gave an almost perfect 9:3:3:1 ratio, the X^2 value being only 0.054. The 1230 plants were 689 *PY*:233 *Py*:232 *pY*:76 *py*.

Fruit Skin Pubescence and Foliage Color—Pp and *Gg* genes. Peach (*ppGG*) by Honor Bright (*PPgg*) produced green-foliaged, smooth-fruited hybrids, which segregated into 148 smooth green (*PG*):57 smooth yellow (*Pg*):53 peach Green (*pG*):16 peach yellow (*pg*) plants, X^2 is here also very low, 0.980 (table 6).

TABLE 6

Summary of data from linkage tests between Pp, and Rr, Yy, and Gg genes.

FACTORS INVOLVED WITH *Pp*		F_2 CLASSES				TOTAL	DEVIATION FROM 10.6 RATIO	X^2	*P*
		PX	*Dz*	*pX*	*pz*				
Rr	observed	288	81	92	37	498	13.8±	3.009	.382
	calculated	280.1	93.4	93.4	31.1		7.16		
Yy	observed	689	233	232	76	1230	3.8±	0.054	>.801
	calculated	691.9	230.6	230.6	76.9		11.47		
Gg	observed	148	57	53	16	274	7.2±	0.980	>.801
	calculated	154.1	51.4	51.4	17.1		5.47		

Genetic Relations between Genes P_rp_r and Rr, Yy, Gg and Cc

Numerous Yellow Pear crosses are the source of data, summarized in table 7, tending clearly, on the whole, to show the independence of the "pear" gene, p_r, from the genes controlling flesh and skin color, foliage color and leaf shape.

Fruit Neck Constriction and Flesh Color—P_rp_r and *Rr* genes. Yellow Pear (p_rp_rrr) was crossed with several varieties with the genotypic formula (P_rP_rRR), as Dwarf Aristocrat, Pink Peach, McMULLENS' Pink, and Honor Bright. The results of these crosses are detailed in table 14. The totalled F_2 classes observed were distributed in the ratio: 698 P_rR:254 P_rr:243 p_rR:80 p_rr. This obviously fits the random dihybrid ratio, X^2 equalling 1.53 and P .682.

Fruit Neck Constriction and Skin Color—P_rp_r and *Yy* genes. Yellow Pear (p_rp_rYY) bred with clear-skinned non-pyriform types (P_rP_ryy) like McMULLEN'S Pink and Pink Peach, produced F_2 phenotypes in the following numbers respectively: 167 P_rY:47 P_ry:62 p_rY:8 p_ry and 292 P_ry : 80 P_ry: 94 p_ry:32 p_ry. These numbers, totalled in table 7, deviate from a 10:6 ratio by 10.2 ± 9.07 and give an X^2 value $= 5.672$ with $P = .132$, the low probability being due not to association of factors but to the low frequency of both clear-skinned classes.

Fruit Neck Constriction and Foliage Color—P_rp_r and *Gg* genes. Yellow Pear (p_rp_rGG) on Honor Bright (P_rP_rgg) yielded an F_2 generation composed of 164 P_rG:44 P_rg:54 p_rG:17 p_rg plants, deviating little from expectation and giving a P value of .536.

Fruit Neck Constriction and Leaf Shape—P_rp_r and *Cc* genes. The Yellow Pear-McMULLEN'S Pink cross brought together from different parents the p_r and C genes. Their F_2 progeny fell into four distinct phenotypes, P_rC, P_rc, p_rC, and p_rc, with the ratio: 166:50:54:16, agreeing excellently with expectation (table 7).

Genetic Relations between Genes *Rr* and *Yy*, *Cc* and *Ff*

The factors for flesh color, *Rr* have been involved in many crosses with those for skin color, and in a few crosses with those for leaf shape and fruit contour. Summarized data from these combinations are given in table 8.

Fruit Flesh Color and Skin Color—Rr and *Yy* genes. These genes have been brought together in many crosses both in the repulsion phase (*RRgg* $\times$*rrYY*) and in the coupling phase (*RRYY* $\times$*rryy*). The findings are set down with details in table 15. Aside from occasional F_2 populations show-

ing a lack of the clear-skinned classes, as noted freqeuntly in other cases, the ratios are in good agreement with the 9:3:3:1 frequencies.

Flesh Color and Leaf Shape—Rr and *Cc* genes. In the cross, McMULLEN'S Pink (*RRcc*)×Yellow Pear (*rrCC*), F_1 plants have cut leaves and red fleshed fruit, and the F_2 plants fall clearly into the four expected groups. The ratio of .163 *RC*:45 *Rc*:57 *rC*:19 *rc*, deviates from the 10:6 ratio less than the probable error, and $P = .642$.

TABLE 7

Summary of data from linkage tests between $P_r p_r$, and Rr, Yy, Gg, and Cc genes

FACTORS INVOLVED WITH $P\ p_r$		F_1 CLASSES				TOTAL	DEVIATION FROM 10.6 RATIO	X^2	P
		P_rX	P_rx	p_rX	p_rx				
Rr	observed	698	254	243	80	1275	19 ± 11.76	1.529	.682
	calculated	717.2	239.0	239.0	79.7				
Yy	observed	459	127	156	40	782	10.2± 9.07	5.672	.132
	calculated	439.9	146.6	146.6	48.9				
Gg	observed	166	44	54	17	279	6.7± 5.38	2.200	.536
	calculated	156.9	52.3	52.3	17.4				
Cc	observed	166	50	54	16	286	3.2± 5.48	0.608	>.801
	calculated	160.9	53.6	53.6	17.9				

Flesh Color and Fruit Contour—Rr and *Ff* genes. Albino (*rrff*) bred with Dwarf Aristocrat or Honor Bright (*RRFF*) produced F_1 plants bearing red-fleshed fruits smooth and regular in outline, and these in turn split up in the following generation into types: red-fleshed, regular or irregular, and yellow-fleshed, regular or irregular. From the first cross the ratio was: 122 RF:42 Rf:39 rF:16 rf, and from the second (Alb.×H. Br):87 RF: 28 Rf: 20 rF:12 rf. The deviation from 10:6 little exceeds the probable error, and $P = .472$. (table 8).

Genetic Relations between Genes *Yy* and *Cc* and *Ff*

Fruit Skin Color and Leaf Shape—Yy and *Cc* genes. Hybrids between genotypes *yy cc* and *YYCC* (see table 16 for details and table 9 for summary of data) gave progeny in four classes with frequencies approximating the 9:3:3:1 ratio, if allowance be made for the deficiency of the clear-skinned terms. The deviation from a 10:6 ratio was 2.04 times the probable error.

Fuit Skin Color and Fruit Contour—Yy and *Ff* genes. Yellow-skinned regular-contoured fruit (*YYFF*) of varieties Dwarf Aristocrat and Honor Bright dominated over the Albino type (*yyff*). The F_2 segregation agreed well with that anticipated from a random redistribution of the two sets of factors. The two inter-variety crosses yielded respectively: 132 *YF*:40 *Yf*: 29 *yF*:18 *yf*, and 78 *YF*:32 *Yf*:29 *yF*:8 *yf*. The mathematical validity of the total numbers as agreeing with expectation is expressed in the constants (table 9).

TABLE 8

Summary of F_2 data from linkage tests between Rr, and Yy, Cc, and Ff genes.

FACTORS INVOLVED WITH *Rr*		F_2 CLASSES				TOTAL	DEVIATION FROM 10.6 RATIO	X^2	*P*
		RX	*Rx*	*rX*	*rx*				
Yy	observed	1026	300	326	96	1742	33.2± 13.5	6.027	.112
	calculated	979.9	326.6	326.6	108.9				
Cc	observed	163	45	57	19	284	4.5± 5.45	1.696	.642
	calculated	159.75	53.25	53.25	17.75				
Ff	observed	209	70	59	28	366	8.2± 6.16	2.554	.472
	calculated	205.9	68.6	68.6	22.9				

TABLE 9

Summary of F_2 data from linkage tests between Yy, and Cc, and Ff genes.

FACTORS INVOLVED WITH *Yy*		F_2 CLASSES				TOTAL	DEVIATION FROM 10.6 RATIO	X^2	*P*
		YX	*Yx*	*yX*	*yx*				
Cc	observed	433	135	117	34	719	17.6± 8.6	7.027	.072
	calculated	404.4	134.8	134.8	44.9				
Ff	observed	210	72	58	26	366	7.2± 6.04	2.307	.518
	calculated	205.9	68.6	68.6	22.9				

Genetics Relations Between Genes *Gg* and *Rr*, *Yy*, *Cc* and *Ff*

In Honor Bright crosses the factor controlling yellowing of foliage, *g*, is involved with those controlling fruit colors, leaf shapes, and fasciation of fruit. The findings are summarized in table 10.

Foliage Color and Flesh Color—Gg and *Rr* genes. Honor Bright foliage is green in new growth but soon turns yellow; and its immature fruits are very pale whitish green until they reach the ripening stage, when they assume a bright red color. The white or yellow-fleshed varieties have somewhat similar immature fruits which ripen "albino" or yellow-fleshed. In the four crosses where the *Gg* and *Rr* genes were involved segregation in F_2 plants was perfectly distinct, but difficulty was encountered because of the late ripening of some of the F_2 plants, particularly the yellow-foliaged segregates. In a few populations the classification could not be entirely completed on this account, even by bringing the fruits indoors to ripen.

Some earlier more completely classified F_2 populations appear to show weak repulsion ratios (see table 17 for details) with crossover values of about 40 percent. The later populations, less completely classified, showed a slight excess of the crossover classes, however, which nearly balances the earlier deficiency. The data cannot be considered properly critical. A back-cross generation now in culture will probably furnish the needed facts.

Foliage Color and Skin Color—Gg and *Yy* genes. With reference to these genes Honor Bright has the formula *ggYY*, and White Apple, McMULLEN'S Pink, Golden Queen, Albino and Pink Peach are *GGyy*. Their F_2 generations fell naturally into the *GY*, *Gg*, *gy*, *gy* phenotypes (table 18). The total ratio was 557:168:168:68, deviating from a 10.6 ratio by 24.4 ± 10.0, and giving an X^2 value of 3.816 and a P value of .286. The genes are, therefore, probably not associated.

TABLE 10

Summary of F_2 data from linkage tests between Gg, and Rr, Yy, Cc, and Ff genes.

FACTORS INVOLVED WITH *Gg*		F_2 CLASSES				TOTAL	DEVIATION FROM 10.6 RATIO	X^2	P
		GX	*Gx*	*gX*	*gx*				
Rr	observed	425	151	142	31	749	12.1 ± 9.0	6.195	.104
	calculated	421.3	140.4	140.4	46.8				
Yy	observed	557	168	168	68	961	24.4 ± 10.0	3.816	.286
	calculated	540.6	180.6	180.6	60.0				
Cc	observed	121	34	42	16	213	3.9 ± 4.72	1.444	.700
	calculated	119.8	39.9	39.9	13.3				
Ff	observed	83	31	23	9	147	1.2 ± 3.94	1.210	.745
	calculated	82.7	27.6	27.6	9.2				

Foliage Color and Leaf Shape—Gg and *Cc* genes. Honor Bright (*ggCC*) × McMullen's Pink (*GGcc*) produced an F_2 distribution of 121 green cut-leaf (*GC*): 34 green potato-leaf (*Gc*): 42 yellow cut-leaf (*gC*): 16 yellow potato-leaf (*gc*). The deviation from the 10:6 ratio is less than the probable error, $X = 1.444$ and $P = .70$.

Foliage Color and Fruit Contour—Gg and *Ff* genes. The F_2 product from an Honor Bright (*ggFF*) × Albino (*GGff*) cross demonstrated the independence of the genes concerned. The observed frequencies of the segregating classes: 84 *GF*: 31 *Gf*: 23 *gF*: 9 *gf*, agree very well with the theoretical frequencies (table 10).

CONCLUSIONS

From the data reported in the body of the paper and in the appendix we are clearly justified in distributing the nine pairs of genes described in six separate and independent groups, constituted as follows:

Group 1. *Dd*, *Pp*, P_rP_r, and *Ss*.
Group 2. *Rr*
Group 3. *Yy*
Group 4. *Cc*
Group 5. *Ff*
Group 6. *Gg*

These groupings must, of course, be considered as tentatively fixed bases for as many linkage groups. They are entirely consistent with all the published data reviewed and represent the most compact and logical interpretation of the facts now available.

SUMMARY

1. The first linkage group established for the tomato contains four pairs of genes, controlling respectively: vine habit (*Dd*, tall *versus* dwarf); fruit skin pubescence and structure (*Pp*, smooth *versus* "peach"); one element of the fruit shape complex (P_rp_r, non-pyriform *versus* pyriform); and type of inflorescence (*Ss*, simple *versus* "compound").

2. Within this group F_2 data indicates that *Dd* and *Pp* are completely (?) linked, and the *Dd* and P_rp_r show 13–20 percent crossing over, *Pp* and P_rp_r 17.5 percent crossing over, and *Dd* and *Ss* 20 percent crossing over. (cf. foot note to p. 393.)

3. F_2 data from 20 different combinations of genes two pairs at a time shows that (a) the genes of the first linked group are independent in inheritance from five other factor pairs controlling: fruit flesh color (*Rr*, red *versus* yellow), fruit skin color (*Yy*, yellow *versus* colorless), foliage color (*Gg*, green *versus* yellow), shape of leaf (*Cc*, cut leaf *versus* "potato leaf"), and fasciation of fruit (*Ff*, regular *versus* irregular).

(b) the five pairs of genes: *Rr*, *Yy*, *Gg*, *Cc*, and *Ff* are also independent of each other, with the possible exception of *Gg* and *Rr*.

4. The results collectively establish reasonably fixed bases for six linkage groups, which suggest in outline the genetic pattern of the tomato.

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APPENDIX—TABLES 11-13

TABLE 11

F_2 data from the cross ddRR×DDrr, showing that Dd and Rr are inherited independently.

VARIETIES CROSSED (see p. 394)	D		d		TOTAL
	R	r	R	r	
Dw. Ar. × Y. Pr.	110	54	40	18	222
Dw. Ar. × Alb.	121	42	43	13	219
Total	231	96	83	31	441
Calculated 9:3:3:1	248	82.7	82.7	27.6	

Deviation from 10:6 ratio = 13.6±6.96

X^2 = 3.734 P = .297

TABLE 12

F_2 data from the cross ddYY×DDyy, showing that Dd and Yy are inherited independently.

VARIETIES CROSSED	D		d		TOTAL
	Y	y	Y	y	
Dw. Ar. × P. Pch.	125	49	44	10	228
P. Pch. × Dw. Ar.	135	52	38	15	240
Dw. Ar. × McM. P.	130	29	50	17	226
Dw. Ar. × Alb.	129	34	43	13	219
Total	519	164	175	55	913
Calculated 9:3:3:1	513.6	171.2	171.2	57.0	

Deviation from 10:6 ratio = 3.4±3.14
X^2= .5134 P> .801

TABLE 13

F_2 data from the cross ppyy×PPYY, showing that Pp and Yy are inherited independently

VARIETIES CROSSED	P		p		TOTAL
	Y	y	Y	y	
P. Pch. × Dw. Ar.	259	99	83	27	468
P. Pch. × Y. Pr.	281	78	95	34	488
P. Pch. × H. Br.	149	56	54	15	274
Total	689	233	232	76	1230
Calculated 9:3:3:1	691.9	230.6	230.6	76.9	

Deviation from 10:6 ratio = 3.8±11.47
X^2= .054 P> .801

TABLE 14

F^2 data from the p,p,rr×P,P,RR cross, showing that Pp and Rr are inherited independently

VARIETIES CROSSED	P		p		TOTAL
	R	r	R	r	
Y. Pr. × Dw. Ar.	109	54	41	18	222
Y. Pr. × P. Pch.	282	90	98	28	498
Y. Pr. × McM. P.	153	61	55	15	284
Y. Pr. × H. Br.	154	49	49	19	271
Total	698	254	243	80	1275
Calculated 9:3:3:1	717.2	239.0	239.0	79.7	

Deviation from 10:6 ratio = 19±11.76
X^2=1.53 P= .682

TABLE 15

F_2 data from the cross RRyy×rrYY, indicating that Rr and Yy are inherited independently.

VARIETIES CROSSED	R		r		TOTAL
	Y	y	Y	y	
P. Pch. × Y. Pr.	300	88	86	24	498
McM. P. × Y. Pr.	170	38	60	16	284
Total	470	136	146	40	782
Calculated 9:3:3:1	439.9	146.6	146.6	48.9	

Deviation from the 10:6 ratio = 21.2 ± 8.77
X^2 = 6.586 P = .088

F_2 data from the cross RRYY×rryy, showing that Rr and Yy are inherited independently.

VARIETIES CROSSED	R		r		TOTAL
	Y	y	Y	y	
Dw. Ar. × Alb.	131	33	41	14	219
H. Br. × Wh. App.	132	39	42	11	224
H. Br. × G. Q.	57	21	22	7	107
R. C. × Alb.	85	31	26	7	149
H. Br. × Alb.	88	27	22	10	147
R. C. × G. Q.	63	23	21	7	114
Total	556	174	174	56	960
Calculated 9:3:3:1	540	180	180	60	

Deviation from 10:6 ratio = 12 ± 10.04
X^2 = 1.141 P = .769

TABLE 16

F_2 data from the cross YYCC×yycc, showing that Yy and Cc are independent in inheritance.

VARIETIES CROSSED	Y		y		TOTAL
	C	c	C	c	
McM. P. × Dw. Ar.	141	39	38	8	226
McM. P. × H. Br.	114	45	37	13	209
McM. P. × Y. Pr.	178	51	42	13	284
Total	433	135	117	34	719
Calculated 9:3:3:1	404.4	134.8	134.8	44.9	

Deviation from 10:6 ratio = 17.625 ± 8.62
X^2 = 7.027 P = .072

TABLE 17

F_2 data from the cross ggRR×GGrr, showing that Gg and Rr are inherited independently.

VARIETIES CROSSED	G		g		TOTAL
	R	r	R	r	
H. Br. × Wh. App.	123	44	48	9	224
H. Br. × G. Q.	57	24	21	5	107
H. Br. × Alb.	90	25	25	7	147
H. Br. × Y. Pr.	155	58	48	10	271
Total	425	151	142	31	749
Calculated 9:3:3:1	421.3	140.4	140.4	46.8	

Deviation from 10:6 ratio = 12.125 ± 8.97
X^2 = 6.195 P = .104

TABLE 18

F_2 data from the cross ggYY×GGyy, showing that Gg and Yy are inherited independently.

VARIETIES CROSSED	G		g		TOTAL
	Y	y	Y	y	
H. Br. × Wh. App.	132	35	42	15	224
H. Br. × McM. P.	129	32	30	18	209
H. Br. × G. Q.	60	21	19	7	107
H. Br. × Alb.	89	26	21	11	147
H. Br. × P. Pch.	147	54	56	17	274
Total	557	168	168	68	951
Calculated 9:3:3:1	540.6	180.2	180.2	60.0	

Deviation from 10:6 ratio = 24.4 ± 10.0
X^2 = 3.816 P = .286